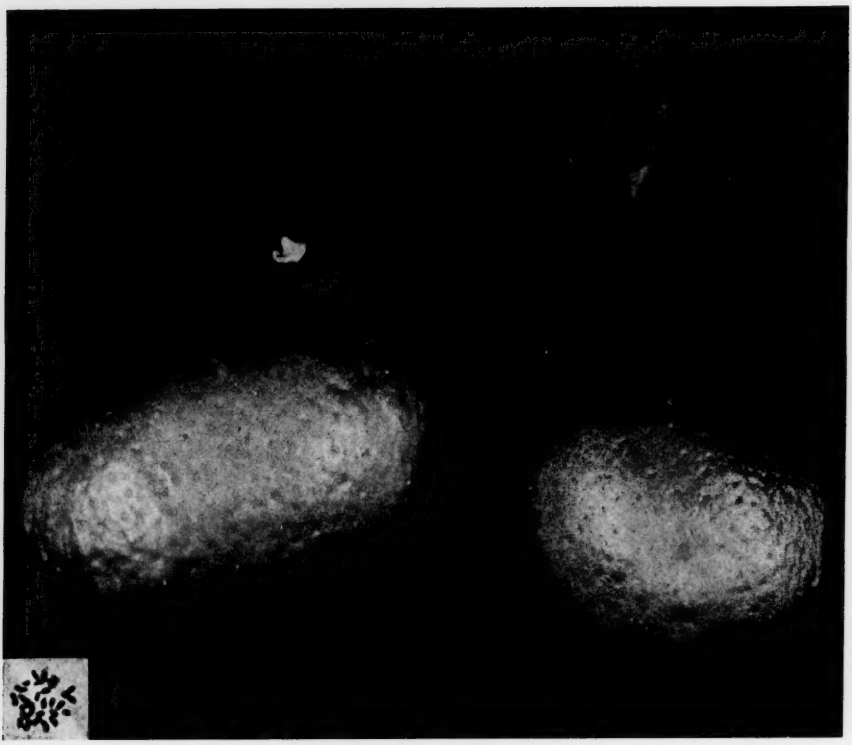
The Journal of HEREDITY



TRIPLOID TAHITI LIME
(SEE PAGE 277)

The Unique Morlok Quadruplets

Twin Studies on Tuberculosis

Maps for Invisible Worlds

VOLUME 34 SEPTEMBER, 1943 NUMBER 9

The American Genetic Association

An incorporated organization devoted to promoting a knowledge of the laws of heredity and their application to the improvement of plants, animals, and human racial stocks

DAVID FAIRCHILD, President Vice-President, SEWALL WRIGHT Secretary, C. E. LEIGHTY
Treasurer, J. H. KEMPTON

EDITORIAL BOARD

Managing Editor, ROBERT C. COOK

M. DEMEREC GEORGE M. DARROW
H. J. MULLER R. R. GRAVES PAUL POPENOE

Members of the Council are ex-officio members of the Executive Board

COUNCIL

W. E. CASTLE, Emeritus Professor of Genetics, Harvard University, and Research Associate of the Carnegie Institution of Washington.
LEON J. COLE, Professor of Genetics, University of Wisconsin.
O. F. COOK, Bionomist in Charge of Breeding Investigations, U. S. Department of Agriculture.
GEORGE M. DARROW, Horticulturist in Charge of Small-Fruit Breeding, Horticultural Field Station, Beltsville, Maryland.
DAVID FAIRCHILD, Agricultural Explorer, U. S. Department of Agriculture.
HARRY V. HARLAN, Senior Agronomist in Charge of Barley Investigation, U. S. Department of Agriculture.
J. H. KEMPTON, Bureau of Plant Industry, U. S. Department of Agriculture.
W. V. LAMBERT, Associate-Director, Purdue University Agricultural Experiment Station, Lafayette, Indiana.
C. E. LEIGHTY, Agronomist in Charge of Dry Land Agriculture, U. S. Department of Agriculture.
HUGH C. McPHEE, Chief of Animal Husbandry Division, U. S. Department of Agriculture, Washington, D. C.
MARCUS M. RHOADES, Department of Botany, Columbia University, New York City.
SEWALL WRIGHT, Professor of Zoology, University of Chicago.

ADVISORY COMMITTEE

W. S. ANDERSON, Genetics Department, University of Kentucky, Lexington, Kentucky.
E. B. BABCOCK, Genetics Department, University of California, Berkeley, California.
E. A. BESSEY, Professor of Botany, Michigan State College, Lansing, Michigan.
A. F. BLAKESLEE, Plant Geneticist, Carnegie Institution, Cold Spring Harbor, New York.
W. E. BRYAN, Professor of Plant Breeding, University of Arizona, Tucson, Arizona.
C. B. DAVENPORT, Carnegie Institution of Washington, Cold Spring Harbor, L. I., New York.
RUSSELL W. DUCK, Livestock Editor, *The Rural New Yorker*, Syracuse, N. Y.
R. A. EMERSON, Professor of Plant Breeding, Cornell University, Ithaca, New York.
C. P. GILLETTE, President, The Genetic Foundation, Fort Collins, Colorado.
C. M. GOETHE, Sacramento, California.
HARRISON R. HUNT, Professor of Zoology, Michigan State College, East Lansing.
H. S. JENNINGS, Professor of Zoology, Johns Hopkins University, Baltimore, Maryland.
D. F. JONES, Plant Breeder, Connecticut Agricultural Experiment Station, New Haven, Conn.
M. A. JULL, Senior Poultry Husbandman, U. S. Department of Agriculture, Washington, D. C.
F. B. LINFIELD, Director, Montana Experiment Station, Bozeman, Montana.
C. C. LITTLE, Director, Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine.
JAY L. LUSH, Professor in Animal Husbandry, Iowa State College, Ames, Iowa.
H. H. NEWMAN, Professor of Zoology, University of Chicago, Chicago, Ill.
T. S. PALMER, Expert in Game Conservation, U. S Dept. of Agriculture, Washington, D. C.
JOHN H. PARKER, Professor of Agronomy, Kansas State College, Manhattan, Kansas.
JOHN W. SCOTT, Professor of Zoology, University of Wyoming, Laramie, Wyoming.
A. D. SHAMEL, Physiologist, U. S. Department of Agriculture, Riverside, California.
GEORGE H. SHULL, Professor of Biology, Princeton University, Princeton, New Jersey.
EDMUND W. SINNOTT, Professor of Botany, Barnard College, Columbia University, New York City. (Representative of A. G. A. on Council of Amer. Assoc. for the Advancement of Science.)
W. W. SMITH, Professor of Animal Husbandry, Purdue University, Lafayette, Indiana.
A. B. STOUT, Director of Laboratories, New York Botanical Gardens, New York City.
W. T. SWINGLE, Plant Physiologist in Charge of Breeding Investigations, U. S. Department of Agriculture, Washington, D. C.
W. H. TOMHAVE, American Aberdeen-Angus Breeders' Association, Chicago, Ill.
H. E. WALTER, Professor of Biology, Brown University, Providence, Rhode Island.
H. J. WEBBER, Prof. Subtropical Hort., Citrus Experiment Station, Riverside, California.
E. N. WENTWORTH, Armours' Bureau of Agr'l Research and Economics, Chicago, Illinois.
P. W. WHITING, Zoological Laboratory, University of Pennsylvania, Philadelphia, Pa.

The JOURNAL OF HEREDITY is published monthly by the AMERICAN GENETIC ASSOCIATION at 32nd Street and Elm Avenue, Baltimore, Md., and 1108 Sixteenth St., N. W., Washington, D. C. Address all communications regarding editorial matters to Room 205, 1108 Sixteenth St., N. W., Washington, D. C. Communications regarding the business management of The JOURNAL OF HEREDITY may be addressed to the publication office, 32nd Street and Elm Avenue, Baltimore, Md., or to 1108 Sixteenth Street, N. W., Washington, D. C. Entered as second-class matter February 24, 1923, at the postoffice at Baltimore, Maryland. Acceptance for mailing at the special rate of postage provided for in section 1103, Act of October 3, 1917, authorized December 20, 1918.

Contents copyrighted 1943 by the American Genetic Association. Unauthorized reproduction of material from THE JOURNAL not permitted. The title "Journal of Heredity" is registered as a trade-mark in the U. S. Patent Office.

$3.50 a Year. 35c a Copy. Prices of back numbers on application.

The

Journal of Heredity

Vol. XXXIV **SEPTEMBER, 1943** **No. 9**

CONTENTS

STUDIES OF QUADRUPLETS—V
By Iva C. Gardner and H. H. Newman 259

TWIN STUDIES ON GENETIC VARIATIONS IN RESISTANCE TO TUBERCULOSIS
By Franz J. Kallmann and David Reisner 269

TRIPLOID VARIETIES OF CITRUS
By C. A. Krug and Oswaldo Bacchi 277

AN ISOLATED HUMAN CASE OF MALFORMED UPPER EXTREMITIES AND THORAX
By Eugene Brown 284

Notes and Reviews

ERRORS IN TURKEY PLUMAGE ARTICLE 257

ONE WORLD? 264

CITRUS CHROMOSOME NUMBERS 283

A QUADRUPLET SET CONSISTING OF TWO PAIRS OF IDENTICAL TWINS 283

Errors in Turkey Plumage Article

Because of a proof lost in the mails and of a simultaneous mishap in the editorial office, a number of errors occurred in the article "The Inheritance of Plumage Color in the Turkey," pages 246-256, the August 1943 JOURNAL OF HEREDITY. Most of these are errors in figure numbering, which in general read one number too low. These corrections are as follows:

Page 246—ninth and tenth text lines from bottom of left-hand column should read: ". . . linked recessive factor. Later[2] he studied an autosomal recessive factor (*l*) producing light down color . . ." Right-hand column, three lines below subhead: 469 should read 369.

Page 247, right-hand column, third line of last paragraph: most should read almost. Fourth line from bottom: (*B*) should read (*Bb*).

Page 249, left-hand column, sixteenth line from bottom: Figure 8*C* should read Figure 10*C*. Right-hand column, first line of first full paragraph, Table II should read Table III. Ninth line below subhead: Figure 9*E* should read Figure 10*E*.

Page 250, left-hand column, seven lines from top: Figure 8*D* should read Figure 9*D*. Second line of last paragraph: Figure 9*D* should read Figure 10*D*. Right-hand column, eight lines from top: Figures 9*F* and 10*B* should read Figures 10*F* and 11*B*. Sixth line of succeeding paragraph: Figure 10*A* should read Figure 11*A*.

Page 252, left-hand column, third line from bottom: Figure 10*E* should read Figure 11*E*.

Page 254, left-hand column, second line under first subhead: Figure 9*D* should read Figure 10*D*. Sixth line under second subhead: Figure 9*D* should read Figure 10*D*. Fourteenth line under same subhead: Figure 10*A* should read Figure 11*A*. Second line from bottom of column: Figure 3*R* should read Figure 11*E*. Right-hand column, second line from top: Figure 10*F* should read Figure 11*F*. Third line from bottom of this paragraph: Figure 10*D* should read Figure 11*D*. Same line and line above: speciments should read specimens, and heerozygotes should read heterozygotes.

Page 256, left-hand column, first line under sub-head: summary should read analysis. Twelfth line lower down should read: ". . . Bronze differs from red by an incompletely dominant autosomal factor (*R*)." Right-hand column, third line from end of text, summarization should read analysis. Literature citations 6-9 inclusive are abstracts.

The editor and the authors greatly regret these errors which were carried through to publication as a result of these simultaneous accidents.

FOUR WITH THE SAME HEREDITY

Figure 1

The Morlok Quadruplets are now over thirteen years old. They are the only set of quadruplets whom careful examination has proved to be identical. Unlike the Dionne quints, the Morlok girls have had a normal bringing up, with very little special care. They have attended public school and taken part in normal activities of children in their community.

STUDIES OF QUADRUPLETS.

V—The Only Living One-Egg Quadruplets

IVA C. GARDNER AND H. H. NEWMAN*

IN a previous contribution to this series of quadruplet studies[6] it was stated that four types of human quadruplets, with respect to zygotic origin, have been identified: one-, two-, three- and four-egg quadruplets. We have already reported on two cases of two-egg quadruplets (the Badgetts and the Kaspars[3,4]), one set of three-egg quadruplets (the Keys[7]), and one set of four-egg quadruplets (the Perricones[8]). In this paper we shall report on and discuss the implications of the only set of one-egg quadruplets that have ever been authentically diagnosed as such. Certainly in our time no one-egg set of quadruplets has survived as a whole set beyond early infancy, and these children represent almost as rare a phenomenon as the Dionne quintuplets themselves.

The Morlok quadruplets suffered the same prenatal, natal and early postnatal handicaps as did the Dionne quintuplets and their survival as a group of vigorous, healthy and intelligent youngsters is only a little less remarkable than that of the quints. They had, moreover, no personal physician, no trained personal nurses or governesses whose sole duty it was to look after their health and education. No commodious home was especially designed and built for them, and no special police guarded them day and night. Yet today they compare favorably with the quints in almost every way and in some ways excel them. If there had been one more of them they would be today as famous as the Dionnes. It is interesting to speculate as to why the addition of one more makes such a tremendous difference in popular interest. The relative rarity of fours and fives is no doubt the main factor, for quintuplets are almost a hundred times as rare as quadruplets, and the Dionnes are the only quintuplets who have ever survived beyond early infancy. Quadruplets are relatively common, there being at least seven complete sets living in the United States at the present time. Yet this much can be said for the Morloks: they are the only unquestionably one-egg set of quadruplets of which we, in extensive and intentive research on multiple human births, have any record. This degree of rarity or uniqueness hardly entitles them to be regarded among "the wonders of the world," but their unique character deserves to be recognized.

From the standpoint of their potential usefulness to science it seems to us that the Morlok quadruplets are of greater value than the unapproachable and closely guarded quintuplets. The parents of the Morlok quadruplets have shown a fine and a commendable co-operative spirit in furthering our studies. There is no type of research involving the use of human multiple sets that could not be carried on as well with a one-egg set-of-four as with a one-egg set-of-five. The number five has no magic properties.

In some ways the Morloks are much more favorable for developmental and nature-nurture studies than the Dionnes. They show a higher level of mental ability, they are better adjusted to normal social life, their language is the same as that in which the standard psychological tests used in many twin studies are formulated. They have not been confused by trying to learn two languages at once to the detriment of both, and they are not wards of a government and therefore somehow sacred.

*As in previous articles by these authors, the data were gathered by Dr. Gardner and were analyzed and written up by Dr. Newman.

Birth and Parentage

The Morlok quadruplets were born May 19, 1930, to Mr. and Mrs. Carl Morlok at the Edward W. Sparrow Hospital in Lansing, Michigan. A prize was offered by a local newspaper for the best suggestion for naming the four babies. The winning suggestion was that they be given names with initials corresponding to the first letters of the four words comprising the name of the hospital where they were born, namely, E., W., S., and H. The parents named them Edna, Wilma, Sarah, and Helen.

Because they were born almost four years before scientific and popular initerest in human multiple births had been aroused by the dramatic advent of the quintuplets, the birth of these quadruplets was only moderately publicized. They were given very little help by the community. At the time of their birth Mr. Morlok was unemployed. He was helped by the city of Lansing to the extent of being given a modest dwelling, rent free. He was subsequently appointed city constable, a position he still holds.

Mr. Morlok was born in Germany and Mrs. Morlok is American born of German-Irish stock. So far as the parents know, there have been no twins or multiple births among relatives of either family, a fact that accords with the present view that one-egg twinning is not hereditary.

Zygotic Diagnosis

Several years ago, on the basis of an intensive study of the finger, palm and sole prints of these quadruplets, J. W. MacArthur and C. T. MacArthur[5] diagnosed them as "monozygotic quadruplets." We have now in our possession a reasonably adequate set of palm and finger prints taken some years later by Dr. Gardner. On of the basis of these prints alone—and uninfluenced by the MacArthurs' prior diagnosis—we would have no hesitation in diagnosing this group of four as a one-egg set. Nothing would be gained by a republication of the prints or their formulation, for prints do not change during the life of the individual except in size. There is no material difference between the formula published by the MacArthurs' and our own.

Further evidence of the one-egg origin of this set of quadruplets, if further evidence be needed, is afforded by the extremely close inter-resemblances in features, expressions, body build, coloring, dentition, and anthropometric proportions. As may be seen in the photograph (Figure 1), even though the transitory expressions are somewhat different, the four girls are almost as much alike as any pair of one-egg twins. Helen is about 5 cm. shorter than the average of the other three, but in other respects is hardly distinguishable from them. In order to tell one child from another it is

TABLE I. Physical Characteristics of the Morlok Quadruplets at 10 Years, 3 Months of Age.

Character	*Edna*	*Wilma*	*Sarah*	*Helen*
Standing height	133cm.	129cm.	132cm.	126cm.
Sitting height	67cm.	66cm.	66cm.	65cm.
Weight	54 lbs.	51 lbs.	53 lbs.	49½ lbs.
Head length	17.7cm.	16.5cm.	17.7cm.	17cm.
Head width	12.5cm.	12.2cm.	12.5cm.	12cm.
Cephalic index	70.6	73.9	70.6	70.6
Eye color	light brown, some flecking	same	same	same
Hair color	light candy	same	same	same
Hair texture	fine, straight	same	same	same
Skin color	fair	same	same	same
Teeth	even, no cavieties	same	same	same
Diseases	measles, mumps	same	same	same

necessary to focus attention upon rather trivial distinguishing features such as differences in hair arrangement or even clothing. Table I gives the physical measurements and other easily observed personal characteristics of the group as they were recorded by Dr. Gardner on August 20, 1940, when the girls were ten years and three months old. Each of the measurements was made three times, with very slight deviations in the repeated recordings, and the average of the three measures is given in the table.

It will be seen from this table that Edna is the tallest and heaviest and that Helen is the shortest and lightest, but the differences in these respects between the two extreme members of the set are no greater than the average between one-egg twin pairs. The head measurements of Edna and Sarah are identical, but Wilma has the smallest head, even smaller than of Helen whose body size is the smallest of the group. The cephalic indices of Edna, Sarah and Helen are identical (70.6), while that of Wilma is 73.9. Eye color and flecking pattern, hair color and texture, skin color, dental characteristics are almost exactly the same in all four. The disease record is the same for all, except that Wilma has had two tonsilectomies and has suffered more from sore throat than the others. Helen has always been comparatively delicate.

On the basis of physical resemblances alone the experienced student of twins would have no hesitation in diagnosing these girls as one-egg quadruplets. This diagnosis is reinforced by that of the MacArthurs based on dermatoglyphics, with which our studies are in essential agreement.

Mental Resemblances and Differences

Three standard intelligence tests were administered to the Morlok quadruplets by Dr. Gardner when they were ten years, three months old. Two of these were mental ability tests: the Stanford Binet (Form L) and the Army Beta. The third was the Stanford Achievement Test. The results of these tests are summarized in Table II. The quads are given arbitrary numbers to avoid invidious comparisons.

Dr. Gardner regards the Stanford Binet test as a more reliable criterion of the mental rating of these quadruplets as compared with other individuals in our population than are either of the other tests, for the Stanford Binet test was administered individually, while the other tests were given to the group. This difference in method of administering the tests should, however, have no effect on the differences exhibited within the group.

All four of the girls were rather slow, deliberate and meticulous. When speed of performance was a factor in scores, all made a rather poor showing as compared with other children of the same age and grade. The Schenze quadruplets, who are to be dealt with in a later paper of this series, made a better showing on the Army Beta and the Stanford Achievement tests than did the Morloks although the Schenses took these tests when they were a year younger than the Morloks. The Morlok girls are doing well in school, having at the age of ten nearly completed the fourth grade. The teachers report that their work is uniformly careful and painstaking and that no one of them stands out from the others in general ability.

According to all of the tests, however, I ranks highest in almost every item, while IV either ranks lowest or ties for lowest in every item. The spread be-

TABLE II. Results of Mental Tests.

Stanford Binet (Form L):	I	II	III	IV
M.A.	11:4	11	11:3	10:5
I.Q.	110	107	109	101
Army Beta:				
M.A.	9:2	9:2	9:2	9:
I.Q.	90	90	90	87
Stanford Achievement:				
Paragraph meaning	9	7	7	7
Word meaning	15	16	9	9
Dictation	80	76	76	70
Arithmetic reasoning	5	3	5	1
Arithmetic computation	15	16	10	9
Total score	124	118	107	96

tween the scores of I and IV is greater than the mean intrapair difference in fifty pairs of one-egg twins reared together and about equal to the mean difference of twenty pairs of one-egg twins reared apart. The differences between IV and II and between I and III are of the same order as those between individuals of one-egg twin pairs.

Correlation Between Height and Weight and Mental Ability

In the rather extensive studies of the Dionne quintuplets by the University of Toronto group of geneticists and psychologists[1] it was obvious that the tallest and heaviest of the quints, Yvonne, was consistently the brightest mentally, while Marie, the smallest, ranked as consistently lowest in this respect. The next to smallest, Emilie, ranked next to lowest, while Annette and Cecile, who are about equal in size and weight, shared second place in mental ability.

This situation is closely paralleled by the Morlok quadruplets. They rank in exactly the same order in height and weight as they do on the Stanford Binet test, which was administered somewhat more carefully than the other tests. On the Army Beta test three of them were equal and one somewhat lower. On the Stanford Achievement test the middle two on the Stanford Binet test changed places. This change of places when different tests were given or when the same test was repeated at different times was observed for Annette and Cecile among the Dionne quintuplets.

The mental differences in either the Dionnes or the Morloks are not large in terms of predicting school aptitude. They are no greater in fact than the changes of five to ten points which often occur when an individual is compared with himself on a retest. The differences between high and low were persistent, however, during the several years over which the Dionnes were studied. In the present study, which is cross sectional rather than developmental, the difference between high and low is consistent on three kinds of tests.

There seems then to exist among one-egg multiple sets a correlation between height and weight differences and mental ability differences. The differences cannot of course be due to differences in heredity, for a set of one-egg multiplets must be genetically identical. Such differences must therefore be in some way environmentally induced. It is well known that pronounced differences in fetal blood supply are common among one-egg twins and doubtless exist among higher one-egg sets. Some members of such sets are favored and others penalized. The result is that members of quintuplet and quadruplet one-egg sets differ greatly at birth in weight and stage of development. Such differences last for years; and though they may become lessened, some may be permanent.

It may well be that retarded mental development induced not by the cultural environment but by handicapping during fetal life, is closely related to retarded physical development, both being two aspects of the development of the organism as a whole. Burks,[2] analyzing data from the monozygotic twins in the Harvard Growth Study, found evidence for "fairly striking relations between shifts in the growth rates of intelligence and certain anthropometric traits—especially iliac width and trunk length". This result suggests that when the optimum developmental pattern of an individual is obstructed during growth, the effects may be very pervasive, altering the development of both body and mind.

Differences in Personality and Behavior

In contrast with other quadruplets we have studied, the Morlok girls are the only children in the family and therefore have no other brothers or sisters to whom they might have to adjust themselves. This makes a study of their group behavior somewhat less complex than it would otherwise be. There is much less of group-of-four behavior as a unit than was evident in other quadruplet sets we have studied and more of a tendency toward normal individual behavior. There are no brothers or sisters for the group-of-four to unite against in defense of group rights.

Within the group, however, there are some well defined cleavages. They tend to associate in pairs. The two larger girls play more often together, while the two smaller tend to keep together in play and in friendships. This pairing may have been initiated by and accentuated by the fact that from an early age the two pairs mentioned have had two separate bedrooms. So there may be nothing very difficult for the psychologist to explain in this group-within-the-group behavior.

It is obvious to any observer that the largest and brightest of the girls is the leader and "boss" in group play and other activities and that the others seem to defer to her and accept her authority. She always acts as the spokesman for the group when they are interviewed. Wilma has the reputation in the family of being the "clown" and trouble maker, for it is usually she who starts the occasional small quarrels or disagreements that arise. Sarah is said to be the "artist" of the group, for she seems to have somewhat greater talent or aptitude for drawing, painting and music than do the others. The smallest of the four is the quietest and least dominant of the group. Her smaller size and less excellent health since babyhood have tended to make her a follower rather than a leader. She is also a little slower in her reactions both physical and mental. She occupies a status in her group quite like that of little Marie among the Dionne quintuplets, where Marie has been characterized as the "baby." If we were inclined to emphasize the personality differences of this interesting group of four genetically identical individuals and to overlook their strong, fundamental resemblances, we might venture to characterize them as the "boss," the "clown," the "artist," and the "baby." Such characterizations would, however, be misleading, to say the least, for these behavior peculiarities are at best superficial and probably have arisen from slight differences in size and vigor rather than from inherent differences in personality.

So far, little has been said about activities of the group as a whole, but such activity is encouraged and enjoyed. They sing together as a quartette and are said by good judges to be excellent in their harmony. They are also being trained to dance as a team and in this they show considerable promise. In contrast with the Dionne quintuplets, the Morloks have never been isolated from the rest of the world. They have attended school like ordinary children and seem to have fully adjusted themselves to the social life of the community. Each of the four girls has her own outside friendships, which of course frequently overlap. One is inclined to believe that it might have been a good plan for the Dionne girls to have had as normal a life as the Morloks are enjoying.

Summary

The Morlok quadruplets, the only surviving whole set of one-egg quadruplets in the United States and perhaps in the world, are potentially as valuable for research as are the Dionne quintuplets. In some ways they are more favorable for such studies than are the much more famous quintuplets, who have gradually become increasingly inaccessible.

The Morloks are unquestionably monozygotic. They show about the same group range with respect to physical, mental ability and personality differences as do the quintuplets. They are somewhat above the average in mental ability. Their group unity of action is less pronounced than in other quadruplets studied; they have split up into two pairs in their play and friendships. They are well adjusted to the social life of their community.

Literature Cited

1. Blatz, W. E., *et al.* Collected Studies on the Dionne Quintuplets. Toronto. 1937.
2. Burks, Barbara S. *Thirty-Ninth Yearbook Nat. Soc. Educ.* Part II: 85-96. 1940.
3. Gardner, Iva C., and H. H. Newman. *J. Hered.* 33(10). 1942.
4. ———. *J. Hered.* 34(1). 1943.
5. MacArthur, J. W., and O. T. MacArthur. *J. Hered.* 28(4). 1937.
6. Newman, H. H., and Iva C. Gardner. *J. Hered.* 33(9). 1942.
7. ———. *J. Hered.* 31:(10). 1940.
8. ———. *J. Hered.* 31(4). 1940.

ONE WORLD?

Some Thoughts on Maps for Invisible Worlds

THE title of Mr. Willkie's book and some of the opinions expressed therein represent a point of view which needs to be given much thought in projected voyages toward the New Horizon and the Four Freedoms.

One obvious aspect of the modern scene is that the world is getting smaller. It took Magellan's crew over a *thousand days* in the early 1520's to do what Willkie did in an Army bomber in *160 flying hours*. Unfortunately the physical world is only one of several worlds human beings live in. Mr. Willkie sighted some of the others but did not explore them, and in some quarters it seems to be the habit to pretend they do not exist. Actually the emotional world, the intellectual world, the biological world, all are quite as important to man, the social animal, as is the world of the five continents and the seven seas. It has become the fashion to map the physical world in lurid colors and strange configurations to simplify the problem of depicting a sphere on a flat surface. Nothing is really being done to map some of these other worlds and to integrate these mappings into the final pattern of the four dimensional (or more) maps which will have to be humanity's guide in the future if we are to go forward into more abundant life instead of backward into quicker and gruesomer mass death. In structuring workable plans for enabling man to live at peace with himself, let alone with his neighbors, we are going to have to go on voyages of discovery to lands no bomber can reach. We are going to have to think fast as well as move fast. We must make daring adventures in understanding human beings if there is ever to be hope that their behavior and relationships on this globe are to be harmonious.

These still ignored areas of mental mapping and emotional integration hold the key to the future much more than the strategic crossroads of air and sea and land travel. It is going to be very easy to hold the Pillars of Hercules, Suez, Capetown and Singapore and Panama in a grasp of steel and yet lose everything. These other intangible strongholds, not yet really mapped, hold the key to a realization of the Four Freedoms on a global basis. Is there any way, in so small and increasingly crowded a world, that any of us (individuals or nations) can long enjoy as a "private monopoly" freedom from disease, or want, or tyranny, or stupidity? The time has come when through increasing control over nature, the human race finds itself in one huge basket. None can long escape unless all do, and that will not be easy. All of our knowledge, all of our gifts of thinking and feeling as human beings, all of our emotional drives must be focused on meeting this surpassing emergency. Mr. Willkie sees part of this truth yet only part. He recognizes that modern medical techniques and industrialized skills must be made available to all people. It is not astonishing that he fails to realize that this of itself cannot possibly be enough. The time-scale and the space-scale have both been so vastly contracted by modern technology that even with an enthusiastic pooling of skills and wills, the future is amply *inter*-complicated.

So Many Sparrows Fall

The principle of interdependence which is characteristic of all human problems is one which usually is overlooked or ignored. Even in physics, simple cause-and-effect relationships are found to be less common than was once thought. "Even a sparrow's fall" does shake the whole universe. Throughout all the living world, the give and take between an organism and its environment (which includes other organisms as well as weather and soil and water), is a multiple dependence held in cohesion

by many interactions. A crude example might be cited in the chain of circumstances which followed extermination of coyotes by the Bureau of Biological Survey. Immediately there was an increase in numbers of ground squirrels and gophers. In a variety of ways these humble creatures interfere with the luxurious survival of the sheep whose safety made necessary the decimation of the coyotes. Since some of these rodents carry bubonic plague and other diseases, the slaughter of predators may even mean a disastrous human epidemic. The reverberations of these changes will affect many other plant and animal species, for every species represents such a vastly inter-dependent pattern of give and take. Our human species is no exception to this rule, though we are smart at passing the buck for a time, and at letting posterity worry about it all, for "what has posterity ever done for us"? As the rate of change increases it will be increasingly difficult to avoid harvesting the whirlwind where we have sown windy generalities and wishful evasions.

Left alone in a state of nature, any species lives or dies as an end-resultant of this vastly complex interplay of mutual dependences. In a human context, our unique faculty of social inheritance as well as of genetic inheritance, gives us a whole universe of new inter-complexities. When the world was kept big enough by primitive transportation, things worked themselves out in various ways for small groups; while the larger world-unit continued as an integration of these isolated ups and downs. When we reduce the oceans to mill-ponds and the continents to beaver lodges in these ponds, then things really do begin to get complicated. Now, speeding communications have thrown the whole globe into an interlocking matrix of shifting goods and ideas and people,—many, many people. This brings with it a stern imperative; through man's endless meddling we have reached the place where the human race is having to decide whether its joint fate will be to sink or swim. Few who admit this truism accept the full implications of this fact, or realize the revolutionary outlooks, the skills, and the daringly conceived expedients, the courage and devotion and integrity which must be harnessed to avoid disaster. The world has grown much too small for any of the meddling half-measures so dear to our hearts. It would seem that the geneticist, the eugenist, and the social psychologist must form an effective alliance if essential parts of this job are to be done.

Human Geometry

The main problems which must be faced are several. Basic and inescapable is the question of numbers. In all the world when Magellan set sail there were fewer than four hundred million people —less than the population of India today. It seems likely that the number had not varied greatly for a very long time. Today our global traveller reviews a world population totalling well over two billion souls, a steady and inexorable increase which began about the time of Magellan's voyage. The increase is greater now than at any time in the history of our race, netting for the whole world about *fifty thousand a day.*

Population is one of those thorny problems like the weather about which there is much talk but little is ever done. Unlike the weather, it is not a matter which can be expected to go on taking care of itself indefinitely. Without taking sides on the thorny question as to what is the optimum population of the world, the proposition stands that on a shrinking frontierless world, surely the time will come when there will not be enough resources to go around. That is no longer a long-range contingency. Many competent experts feel that the optimum has already been passed; if not, that it soon will be. One of these experts recently pointed out that it is a simple matter of arithmetic to prove that if the death rate of India were to be reduced to that of England in recent years, the birth rate remaining unchanged, by the end of a century there would be twelve billion people living in India. That would be some six times the present population of the globe. Even those who ridicule

the possibility of overpopulation would be appalled by living in such a world. In saying this we cast no aspersions on the qualities of the people of India. Twelve billion of anybody's supermen offers problems of survival, let alone of decent subsistence, that in the end could only be solved the old stupid cruel way —war, famine, and pestilence.

Puerto-Rican Progress

We have in our own front yard an outstanding example of how our genius for misplaced meddlesomeness works out in practice. In 1898, amidst a glittering display of July Fourth oratory, we freed the people of Puerto Rico from an intolerable tyranny. Surely this worthy venture was undertaken with a wholehearted desire to help the people of Puerto Rico achieve the more abundant life. What has happened in the intervening years is so complex that the total answer has never emerged, but as far as the Puerto Rican people are concerned, the result has been disastrous. In 1934 a Commission was created by the legislature to deal constructively with the tragic crisis which beset the island. More important than "acts of God" such as the periodic hurricanes, or political chicanery and trade discrimination, is the inescapable and perennial fact that the population of the island has more than doubled since we took over, being now more than two million souls. "The United States, having provided just enough minor health improvements to insure more births than deaths, has unwittingly encouraged the survival of the unfittest to an extent scarcely equalled in the entire civilized world. Hungry and indifferent, ill and ignorant, the peon wanders about begetting himself. . . . . He brings these offspring not only into a fixed economic environment where their opportunities will be precisely what his have been, but almost surely into the clutches of malaria, filaria, syphillis, hookworm, tuberculosis, to say nothing of malnutrition, which they can count on definitely." That is what Dr. Eric Matsner found in 1937.

The "Reconstruction" Commission concurred in finding population growth was the primary disaster which must be dealt with before there could be a hopeful future. If the present rate of population increase continues for a century, this tiny island, only a hundred miles long, will have on it as many people as all the continental United States today. We have here a gruesome example of how meddling half-measures, conceived in a spirit of vague good will and an earnest determination to avoid coming to grips with fundamentals, end in creating tragedy and despair. When Mr. Willkie sees the urgent need for Public Health measures in crowded areas, does he also see that these measures are doomed to be worse than useless unless along with the Four Freedoms goes a fifth basic freedom—the right to have only wanted children, and the correlative right of human society as a whole, not to be burdened with a mob of reproductive casuals, intent only on feeding and breeding? "Natural causes" take care of *that* the hard way. To suspend these ruthless checks and to substitute nothing better is not only stupid, it is suicidal and in the end frightfully cruel.

Social Parachutes Needed

It is no answer to this position to say that at the moment we do not know how to control such matters effectively. Along with shorter working hours and Public Health measures which are being promised to all the world, we must develop techniques which will be an answer to the problem of overpopulation. It is nonsense to hope that matters will take care of themselves. They have not in Puerto Rico and they show no prospect of doing so in any of the other acute pressure-areas. It is cruel and stupid to beg the question with such pious hopes. If we get out and let "natural causes" take over, the problem would be solved the merciless way of mounting death rates. It is nonsense to pretend hopefully that this grim reckoning will be magically averted in the long run. Without effective action, disaster is inevitable. If we cannot put Puerto Rico in order, is it not madness to talk of a

worldwide miracle? Would it not be more intelligent to work out first a humane and decent and effective solution of Puerto Rico's problem before we tackle over half the human race? And if there is not time for that, should we not at least profit from the Puerto Rican failure, and pledge ourselves to avoid greater disasters elsewhere?

It is the fashion in certain quarters to point hopefully to new agronomic techniques as offering the possibility of feeding indefinitely expanding populations. As Fibonacci proved in 1202 (when he had to introduce arabic number-theory into Europe to deal with the descendants of a single pair of fecund rabbits) the explosion of unimpeded reproduction must always outrun resources. It is equally folly to attempt to placate the blind god of reckless numbers by a degradation of diet. We go off meat to soy beans; and then to rice, and still there is not enough. In the early years of this century Java was one of the great rice-exporting centers of the East. For the last few years before the Japanese invasion, the redundant population, then over 44 million and increasing a million a year, had absorbed all the crop, and rice was actually being brought from less favored agricultural regions to feed the Javanese. Within a generation Java could have 88 million people instead of 44, and where would the rice come from to feed them? Fibonacci's series posed that problem seven hundred years ago, and the time has come when we must find ways to take his lesson to heart and do something about fecundity in a global context.

It is not a rational answer to allege hopefully that the entire population of the globe can somehow be, by magically waving an economic wand, transferred into an economy of plenty, and that there would then be no problem of overpopulation because fattened people don't beget. Even in the United States where the colonial pattern of many births and deaths has, over a century, given place to one of few births and fewer deaths, the change has taken several generations. We do not even know that in a different culture context the result would be the same. In China, the ideal of a large family seems to be basic to the philosophy of the people. Suddenly to bring this consummation within the range of millions of people might well result in a simply fantastic multiplication of population before economic over-fattening had its way if it could ever be achieved. We know from the Puerto Rican experiment what a little preventative medicine will do to foster population growth in an unpromising situation. The same technique applied globally could easily spell quick disaster rather than ultimate progress.

All of One Blood, But—

And that is only one of the problems. We are in urgent need of facing the reality that the habitable globe is peopled by a number of different kinds of human beings. It makes no difference whether we call these groups "races" or by some other name. Just a brief survey of the news and the discussions going on about world affairs makes it perfectly obvious that these differences bulk large in developing intergroup tensions and antipathies. The cultural anthropologists who loudly proclaim that such differences are purely artificial and that we are all identical twins under the skin, are almost as unrealistic as the stupid racists of the Nazi type. Human differences are a problem that must be recognized and studied and dealt with by understanding and tolerance, not by hush-hush culminating in race riots. Magellan's sailors saw races they never dreamed of, and today those racial groups are our neighbors. What are we going to think about that and do about that? Only the human ostrich can pretend the problem does not exist and that a courageous and realistic and humane attack on it can long be delayed with impunity.

To build this dreamed-of postwar world, it is essential that we and other people learn to know the other side of the world as well as that on which we live. We can hardly escape the destiny which links us closely to Western Asia where half the human race lives today. For Willkie to converse not only with

Generalissimo and Madam Chiang Kai-shek, but also to have a quick glimpse of the Chinese Army was an excellent begining. Obviously it is not nearly enough. Will the millions of American boys who are soldiering at the ends of the earth have the time and the reserves of energy to gain a sympathetic understanding of other people? Far too often it works the other way; Allies fall out merely by rubbing elbows. The good-will booklets which the Army has prepared to help our soldiers understand their neighbors are a small step in the right direction. These books bear about the same relation to the total context as a Bible in the blouse pocket does to preserving a soldier's life against machine gun and artillery fire. Yet, without a sympathetic understanding of the peoples of the world, how can we hope to do the job of leading all mankind to the Promised Land, which we talk so hopefully about?

No Fooling

Implementing even the changes in habits of life which Willkie sees as essential offer a tremendous problem. We run directly into the paradox of means and ends. Man is a socialized creature; part of whose individual personality depends on his biological inheritance. But the way in which this necessary substrate is sculptured by the culture pattern in which man grows up is just as important to the realization of the good life as the biological endowment. Our fascist enemies have given us an arresting example of the impossibility of ever building a decent human culture on biological conceptions which might, if physiology were the whole story, be excellent. A race of physical and intellectual supermen, morally and spiritually dehumanized, would only be super-beasts. The wages of cynicism are cultural death: no human culture which does not base on deeply grounded convictions can long survive. It will be impossible to bluff and fool the people of the world into the New Jerusalem. And ill-conceived half-measures which through folly or fear or political expediency ignore the basic problems of our time can only achieve failure. We will be able to use the hypocrisy of the Nazi high command as a weapon against them if we do not fall into the same pit ourselves. Postwar blueprints which are defective in ignoring the basic realities are in their way as cynical and hypocritical as some of the Nazi effronteries. We doom ourselves if we fall into that error.

The politicians and the statesmen and the formulizers and verbalizers cannot ever develop solutions of this problem of living together as neighbors. It will take all the knowledge we have of anthropology, of sociology, or phychology, and of genetics, and even then a happy solution will be something of a miracle of inspired cooperation and of untapped resources of goodwill. The miracle will not just happen—it will have to be brought about by courageous honesty, faith, and works.

In our voyage toward the Four Freedoms we face vast and practically unexplored areas of group thinking and feeling, of national and individual motivations, which must be mapped before this shrinking world can ever be a safe or hopeful home for all of humankind. To attempt a solution of global problems purely by treaty or by force or by good wishes, or by economic expedients, is doomed to fail. The major problems which confront us at the moment have arisen because man has boldly undertaken to meddle with life and death through technology and biology and medicine. The job is half done and he stands at a portentous crossroads: he either has to give up the battle and go back to incantations and inevitable failure, or he has to finish what he has begun. Magellan did not try to sail unknown oceans blindfolded 500 years ago We would be well advised to use all our skills and all our wisdom and humanity in this difficult time ahead. Charts as well as charters we must have, and to the 1943 Magellans who first circle this globe of the mind and spirit of mankind we must look for the maps without which our shrinking world will inevitably explode in a shattering catastrophe. These basic facts cannot be ignored if our 1943 dream of world unity is to be achieved.—ROBERT COOK.

TWIN STUDIES ON GENETIC VARIATIONS IN RESISTANCE TO TUBERCULOSIS

FRANZ J. KALLMANN AND DAVID REISNER*

IN the study of genetic factors which may play a role in the development of tuberculosis, one must bear in mind that the effects of hereditary and non-hereditary agents in the origin of human morbid conditions cannot be expected to operate in a mutually exclusive fashion or to interact in a simple manner.

Regardless of the type of inheritance involved, modern genetic research has discarded the belief that the part played in the production of a trait by hereditary elements (genes) precludes variations in the manifestations of the trait as a result of specific environmental influences. Likewise there is no reason to infer that the existence of one or more causative agents of a non-genetic nature excludes heredity as a definite factor in the causation of a disease. As is true of any non-pathological trait exhibited by an organism, in relatively common inherited disorders the co-acting roles of heredity and environment may be as essential as "fuel and oxygen are necessary for making a fire" (Woodworth[31]).

The possible significance of hereditary agents in the etiology of tuberculosis, therefore, should not be minimized merely because exposure to infection is known to be one important factor in the production of the disease. The practical result of this might simply be a depreciation of pathogenetic knowledge rather than a reemphasis of the basic role played by the tubercle bacillus in the accepted scheme of causation. On the other hand, any success in the identification of genetic factors which modify the individual vulnerability to the infectious agent would be of very little value without a correct understanding of those specific biological conditions which classify tuberculosis as an infection and imply that the acquisition of a certain number of bacilli is a necessary prerequisite to the initiation of the disease.

The problems caused by this intricate interaction between infectious agents and the genetically determined resistance factors of the individual are further complicated by various uncertainties concerning the type and specificity of the hereditary mechanism or mechanisms which may be involved in the development of tuberculosis. Genetically a fundamental distinction is made between that specific form of susceptibility determined by a single morbid gene having a large effect, and the varying minus variants of a multi-factorial genetic mechanism which result in different degrees of susceptibility to a disease by reason of deficiencies in normal resistance. When many genes interact to produce a graded character such as resistance to infection, their individual effects will be neither potent nor pathogenic, and their interaction will usually be in the direction of a normal state, which in tuberculosis will be a satisfactory state of protection against tubercle bacilli. A pathologic state will be produced by these genes only if they are represented by poorly assorted alleles in the genotype; or if, because of some somatic interference, the genetic potentials fail to emerge effectively in the phenotype.

A single morbid allele may be dominant or recessive in the manner of its inheritance, and it may be completely or incompletely penetrant in its expression;

*From the Division of Research in Genetics of the New York State Psychiatric Institute and Hospital and from the Bureau of Tuberculosis, New York City Department of Health, New York, N. Y. Based on material which was presented in part at the thirty-eighth annual meeting of the National Tuberculosis Association, Philadelphia, Pa., May 6-9, 1942, and published in full in the *American Review of Tuberculosis*, Vol. 47, June, 1943. The study was supported by grants from the Carnegie Corporation of New York and the National Tuberculosis Association.

but the main implication of such a single factor for a specific susceptibility is that an individual cannot develop the disease without being a carrier of the particular allele. Since it is commonly held, however, that no human being is absolutely immune to tuberculosis, it would follow in the case of single-factorial inheritance as assumed by Diehl and Von Verschuer,[8] that there must be two distinct forms of tuberculous disease; namely, an inherited form developed under ordinary conditions of exposure by persons carrying the specific allele, and a non-hereditary form produced in any individual exposed to an overwhelming infection.

The assumption of two different forms of tuberculosis would not be necessary, if variable resistance to the disease were the result of multi-factorial inheritance. The existence of a heredo-constitutional defense mechanism resulting from the interaction of many genes would suffice to explain why the expression of heredity in tuberculosis seems to be of a graded character, ranging from the greatest to the least possible degree of resistance. Since the inherited capacity for the development of high resistance cannot be thought of as providing more than a relative degree of immunity, it is not difficult to see, in terms of multi-factorial inheritance, that the protective system of even a highly resistant person will break down if subjected to massive infection or repeated inoculations.

There is still another complicating feature which is to be considered in the genetic analysis of tuberculosis. Theoretically it is possible that hereditary effects may be associated with the operation of different genetic principles at various stages of the tuberculous process. It may be necessary, for instance, to make a distinction between varying degrees of resistance to the invasion of the tubercle bacillus and varying degrees of resistance to the spread of the established disease. If it could be shown that there actually exists a genetically determined inability to resist tuberculous infection as well as an innate inability to resist the progressive disease, then two such genetic mechanisms—distinguished as "Anfälligkeit" and "Hinfälligkeit" in the German terminology—cannot be assumed to be identical without conclusive biological evidence of their identity.

The need for a thorough clarification of this possible interplay of genetic and non-genetic factors in the determination of variable resistance to tuberculosis is emphasized by a number of clinical and experimental observations. The clinical evidence in support of heredity includes the generally accepted fact that variations in epidemiologic and other environmental conditions alone are unable to explain the consistent differences in tuberculosis morbidity and mortality among certain racial groups.

There is also the common experience that exposure to infection, although basically important in the causation of the disease, often does not suffice to produce manifest tuberculosis. Especially significant in this respect is the failure of many marriage partners of tuberculous patients to develop clinical disease despite most intimate and prolonged exposure. On the other hand, manifest tuberculosis is frequently found in the absence of any known contact history, even if careful inquiries are made to determine the actual source of infection. Evidently, however, although these and other observations indicate variability in individual susceptibility or resistance to tuberculosis, no one of them is by itself sufficiently conclusive to substantiate the theory of a genetic basis for this difference.

More pertinent in this respect are data obtained by experimental methods, especially the findings observed in animal experiments and in twin studies. There may be certain objections to a direct comparison between the pathogenetic conditions of spontaneous human infections and those artificial infections produced in inbred animal stocks. To some extent one may also be justified in questioning the statistical accuracy and genetic interpretation of certain earlier twin investigations. It may even be debatable if it is quite correct to

classify the unqualified twin study method as "experimental," since natural or social forces rather than human ingenuity provide in twins an opportunity to analyze the relative effects of heredity and environment. Unquestionably, however, the results obtained by the different methods of approach in twin studies and animal experimentation correspond closely in demonstrating the significance of heredo-constitutional factors in the determination of variable resistance to tuberculosis. It cannot be denied, therefore, that the execution of careful control investigations is definitely indicated and, at the present state of limited genetic knowledge, rather urgently needed.

Among the animal experiments which have been indicative of genetically determined variations in resistance to tuberculous disease, Lurie's work[14,16] in inbred rabbit stocks, continuing the pioneer studies of Lewis and his associates at the Phipps Institute, deserves particular attention. Consistent inbreeding of ordinary rabbits led to the development of three distinct family groups; namely, a highly resistant family displaying a localized, chronic, ulcerative form of pulmonary tuberculosis analogous to the human reinfection type; several non-resistant families with rapidly progressive, primary, generalized disease; and some moderately resistant families showing various intermediate forms of the disease. Lurie concluded, therefore, that the "resistance of rabbits to naturally or artificially acquired tuberculosis is largely controlled by their genetic constitution, although it may be overwhelmed by large doses of tubercle bacilli." The main variant in the type of disease developed by the three different family groups was seen in the extent of localization of the infection at the portal of entry.

These consistent variations in the inherited resistance of rabbits were found by Lurie to be closely related to the intensity and rapidity with which local immunity was acquired by the tissues of the respective families. Other varying factors revealed as being associated with the two extreme degrees of resistance were high or low skin permeability, rapid or slow development of allergic sensitivity, and intense or tardy production of antibodies.

It is the mononuclear cells which in highly resistant rabbits are assumed by Lurie to possess the capacity for inhibiting the growth of the bacilli to such an extent as to prohibit their multiplication in organs focalized by lymphogenous or hematogenous routes. There is a fulminating progression of the primary focus in non-resistant families, as their mononuclears are unable to interfere with the growth of the bacilli in the cytoplasm. No one of the rabbit families of intermediate resistance is said to have shown such extreme variations in the ability or inability of their mononuclear cells to develop inhibitory properties against the multiplication of bacilli in their cytoplasm.

Whether or not Lurie is correct in suggesting that "the sum total of numerous determinants controls hereditary resistance to tuberculosis in rabbits," and that "the disease types observed in rabbit families of varying inherited resistance are essentially analogous to the different types of tuberculosis developed by different human ethnic groups," the general genetic significance of his findings in rabbits would be unquestionable if similar inherited variations in resistance to tuberculosis could be shown to exist in spontaneous human infection. In order to carry out these essential studies, the only "experimental" method directly applicable in man is the twin method. The comparison of unselected groups of one-egg and two-egg twin pairs provides a truly controlled situation which is indispensable for an objective analysis of the variable effects of genetic potentialities in human tuberculosis.

The effectiveness of the twin method rests upon the distinct hereditary differences between the categories of monozygotic (identical) and dizygotic (fraternal) twins. Monozygotic twins occur as the result of the ability of an embryo to divide at an early stage of development. Since both halves are

derived from a single fertilized ovum each of these embryonic primordia carries precisely the same complement of genes. Such twins are always of the same sex and are genetically as identical as the right and left halves of a single individual. Frequently they resemble each other so closely that it is practically impossible to tell them apart.

Dizygotic twins arise from two separate ova which happen to mature and to be fertilized at the same time. They are genetically no more alike than any other pair of brothers and sisters who are born at different times (siblings). Of course, they may be of the same or of opposite sex. Their genotypes cannot be expected to be composed of the same alleles more frequently than are those of ordinary brothers and sisters.

Under usual conditions, both types of twins are likely to share about the same physical and nutritional environment. Consequently, if an intra-pair difference is greater for dizygotic than for monozygotic twins, the fundamental cause for this difference must be sought in heredity, although it is to be expected, as Woodworth[31] has put it, "that environmental factors may accrue to heredity as interest accrues to legacies of different amounts."

The usefulness of the twin method naturally depends upon the possibility of distinguishing reliably between the two categories of twin pairs. The safest procedure to classify twin partners in respect to their zygotic origin is the modern similarity method, since it is known now that not all of the monozygotic twin pairs are necessarily monochorial. If twinning occurs at a very early stage of development, two separate chorions may be developed even by monozygotic twins, although it is the rule only for dizygotic twins.[17]

The common objection, that a difference in tuberculosis morbidity between one-egg and two-egg co-twins (partners of twin index cases*) might be adequately explained by a more intimate contact in monozygotic twin pairs, is valid at best for twin studies which are numerically limited and open to errors from unrepresentative sampling. In statistically representative surveys this argument can be met by sampling suitable sub-groups, especially by comparing twins of the same and of opposite sex, twins reared together and reared apart, and twins with and without exposure to a common source case. While any intra-pair difference between monozygotic twin partners must be attributed to variations in external or dispositional influences, an intra-pair difference which is greater for opposite-sexed than for same-sexed sets of dizygotic twins may be due either to genetic or non-genetic factors associated with the difference in sex.

This scope of the original twin method can be broadened still further if the number of genetically dissimilar sibship groups reared under comparable environmental conditions is extended to include full siblings (with two parents in common), half-siblings (with one parent in common), and step-siblings (not related by blood) of the twin index cases. Such a combination of a twin and family survey has been adopted in this study and may be called "Twin Family Method." It provides six distinct categories of sibship pairs; namely, monozygotic twins, dizygotic twins of the same sex, dizygotic twins of opposite sex, full siblings, half-siblings, and step-siblings. If the assumed genetic mechanism exists and the part played by the twinning factor is negligible, the statistical expectation will be that the concordance† rates for dizygotic twins should closely correspond to the morbidity rates for full siblings, but should distinctly differ from the rates for the other sibship groups.

*The term "index case" is used in this study in preference to, but synonymously with, the more customary genetic term "proband." It designates those original cases in a family survey, which constitute the starting point of the study and, therefore, must be proved representative of the type of trait-carrier whose relatives are to be investigated as to the recurrence of the trait under observation.

†Similar behavior of two twins in regard to the trait under observation.

Other Twin Studies

Of course, such a detailed analysis requires an extensive and unselected material, while most of the previous twin investigations have been limited either numerically or methodologically. The majority of these studies were case reports of single twin pairs which happened to be concordant or discordant as to certain clinical aspects of the disease, and the few remaining ones were either restricted as to the size of the material or questionable as to the criteria of selection used. Nevertheless, their findings seemed to confirm what had already been suggested by the genealogical investigations of Weinberg,[29] Riffel,[19] Doyer[10] and others; namely, that the closest blood relatives of tuberculous patients tend to develop clinical tuberculosis more frequently, and even not uncommonly in a more similar form, than is observed in members of the general population under comparable conditions of exposure.

Of the individual twin cases so far reported in the literature, the pairs of Bauer,[4] Kretschmer,[13] Czarnecki,[7] Kettelkamp and Stanbro,[11] Klein[12] Siemens,[23] d'Almeida,[2] Schäfer,[22] and Valdés-Lambea[27] seem to have been monozygotic and concordant; those of Alons,[3] Rössle,[21] Czarnecki,[7] and Stamm[25] were probably dizygotic and concordant; and that of Curtius and Korkhaus[6] was later diagnosed by Diehl and Von Verschuer[9] as dizygotic and discordant. The cases described by Ahfeld,[1] Rivers,[20] and Orgler[18] were so incomplete as to the data on zygosity that they cannot be classified at all. The reports of Schokking[24] on nine pairs and of Bossik[5] on 117 pairs were confined to observations in children. The main conclusion of Bossik's[5] survey, which was conducted at the Maxim Gorky Medico-Biological Research Institute in Moscow, was that hereditary factors play a part in susceptibility to infection and in the reaction of the host to the microorganism, since the monozygotic twins showed simultaneous occurrence and similarity in the course of primary tuberculosis more frequently than did the dizygotic twins.

Of the two remaining twin studies dealing with clinical tuberculosis in adults, that of Uehlinger and Künsch[26] was based on 46 pairs, of which 12 were classified as monozygotic and 34 as dizygotic. Concordance as to tuberculosis was observed in 58.3 per cent (seven pairs) of the one-egg sets and in 5.9 per cent (two pairs) of the two-egg sets. Discordance as to tuberculosis was found in five monozygotic and 32 dizygotic pairs, while similarity in environment was reported in seven of the 12 monozygotic pairs and in 25 of the 34 dizygotic pairs.

More conclusive were the results obtained by Diehl and Von Verschuer[8, 9] in a survey which may be rated as statistically representative, although it included only the twin partners of the index cases and was not quite consistent in the selection of cases. Their twin material originally consisted of 127 pairs, but later was enlarged to 205 pairs. Of this total number, however, only 134 sets in the final material and 77 sets in the original material were unselected cases of active ("progressive") clinical tuberculosis ("exclusive of healed primary infections"). In a separate analysis of these various groups the difference in concordance as to tuberculosis between one-egg and two-egg twin pairs ranged from 25 to 70 per cent for the first series of 77 unselected pairs, from 21 to 60 per cent for the total group of 134 unselected pairs, and from 25 to 65 per cent for all 205 pairs studied. These consistent and statistically significant variations in the concordance rates of the two categories of twin partners were shown to be due to the genetic rather than environmental differences between monozygotic twin pairs. The authors concluded, therefore, that hereditary factors play a decisive role in both the origin and the clinical course of tuberculous disease.

The genetic mechanism involved was assumed by Diehl and Von Verschuer to be definitely specific and "most prob-

ably not of a multi-factorial nature," while its modifying effect on the "natural resistance to tuberculous infection" was found to become more pronounced with increasing age. No final attempt was made to analyze the given type of inheritance or the morphological aspects of the inherited factor for susceptibility to tuberculosis in any particular details.

All of these observations indicate the significance of genetically determined variations in susceptibility or resistance to tuberculous disease. The fact remains, of course, that tuberculosis is an infectious disease caused by a specific microorganism and, therefore, modifiable by any environmental conditions, biological and socio-economic, which may influence infections in a given population. Nevertheless, it is necessary to clarify the important part which seems to be played by genetic factors in the variability of the pathologic effect produced by the tubercle bacillus. At present, no better method is available for this purpose than the "Twin Family Method."

II. The Organization of the Study

In order to obtain statistically representative material for the application of the Twin Family Method, the study was organized on a long-term, state-wide basis. The twin index cases were collected from the resident populations and new admissions of the tuberculosis hospitals as well as from the patients of the tuberculosis clinics in the State and City of New York. The danger of bias on account of major technical selective factors in the sampling of the material was avoided by referring the determination of the index cases to the staffs of the hospitals and clinics cooperating in the survey. Their collaboration was procured through the courtesy of the respective departmental executives in the New York State Department of Health and the New York City Departments of Health and Hospitals.

In the procedure of collecting the twin index cases, no attention was paid to the survival, health status or availability for personal examination of the twin partners. This precaution was necessary in order to avoid an excess of cases whose twinning status had become known because of conspicuous features in their concordance as to tuberculosis or any other finding of special clinical interest. Another precautionary measure consisted of excluding from the statistical survey proper those twin cases which had not been reported through official channels. The only criteria for selection of the index cases were, therefore, that they be born by multiple birth and that they had come under observation of one of the cooperating agencies with a diagnosis of tuberculosis. Accordingly, residence of one member of a set of twins in an institution located within the State of New York, rather than legal residence of a twin patient or his family in this state, served as the accepted standard in the limitation of the material.

The diagnostic aspects of the study were strictly a matter of the investigators' research, since it was advisable to ascertain the clinical status of cases beyond reasonable doubt and in accordance with a uniform classification. Whenever possible, therefore, the diagnosis of both tuberculosis and zygosity was made on the basis of personal examination and extended observation. The clinical data required for a diagnosis of tuberculosis included complete histories, chest X-rays and sputum examinations. No information essential for clinical or genealogical classifications was ever accepted without substantial evidence secured by personal visits, regardless of whether these data were obtained from hospital records, the patients themselves or any of their relatives. Nor was it consistent with the rules of the investigation to classify any person included in the survey as free from clinical disease without the availability of current negative roentgenograms and a detailed history of exposure. Systematic efforts were also made in every case to collect the birth records

and childhood photographs of the twins, although the final twin diagnosis was always based on the personally obtained results of the similarity method rather than on descriptions, impressions, or the character of the fetal-membranes.

The practical efficiency of this system is demonstrated by the total number of 657 twin cases reported within a period of less than five years, and by the distribution of monozygotic and dizygotic sets in the total group of 308 twin pairs used in the statistical analysis of twin data on reinfection tuberculosis (Part III). There are 78 monozygotic and 230 dizygotic twin pairs with manifest reinfection tuberculosis in at least one member or, more precisely, 308 pairs constituted by 616 twins, of whom 334 were index cases affected by reinfection tuberculosis (Table 1). Of the 230 two-egg sets, 118 are same-sexed and 112 are opposite-sexed.

The random sampling of this material is indicated by the close correspondence between the statistically expected figure of 25.6 per cent for the proportion of monozygotic twin pairs in an unselected American twin group, and the corresponding figure of 27.3 per cent as calculated according to the Weinberg Differential Method for the material of the present study. The main principle of this method is that there should be equal numbers of same-sexed and opposite-sexed twin pairs in the absence of one-egg pairs. Therefore, in multiplying the number of boy-girl pairs by two and subtracting this product from the total number of pairs under observation, that is, 308 minus 224 in the present instance, one obtains that comparable figure for the proportion of monozygotic sets which should, and in this material practically does, correspond to the rate expected in a general population group.

The ratio of white to non-white twin index cases is about 5:1 and is of significance in this analysis mainly because it was taken as the basis for computing a corrected morbidity figure valid for a general population with comparable age and racial distributions. The need for such a statistical correction is evident in a survey which requires comparison of collective morbidity rather than age-specific mortality rates, although it includes genealogically diverse kinship groups such as parents and siblings of the index cases.

It is shown in Table II that the fact of being the parent of a tuberculous index case practically eliminates this person's chance of dying from tuberculosis before the age of twenty-five, while the siblings of the index cases reach the peak of their tuberculosis mortality between the ages of twenty and twenty-four. Obviously, the morbidity figures for such heterogeneous relationship groups, collected through index cases affected by tuberculosis or any other trait not manifested at birth, cannot be directly compared either with each other or with a general population rate, unless they are corrected for major differences in age distribution by the same statistical method. In fact, the uniformity of this corrective procedure may be considered

TABLE I. Distribution of age and sex in the twin index pairs.

AGE GROUP	TWIN INDEX CASES					TWIN PARTNERS				
	Living		Dead		Total number	Living		Dead		Total number
	Male	Female	Male	Female		Male	Female	Male	Female	
15-19	10	6	2	4	22	5	10	—	2	17
20-24	26	44	13	9	92	30	44	5	2	81
25-29	18	36	10	20	84	18	34	1	5	58
30-34	21	21	3	6	51	18	22	—	1	41
35-39	12	10	6	7	35	11	17	1	1	30
40-44	9	8	2	3	22	8	13	1	1	23
45-49	2	4	3	3	12	8	4	—	2	14
50-54	2	2	2	3	9	5	4	—	—	9
55-59	—	1	3	—	4	3	1	—	1	5
60-64	—	1	2	—	3	3	—	1	—	4
Total number	100	133	46	55	334	109	149	9	15	282

TABLE II. Tuberculosis mortality in the siblings and parents of the index cases.

AGE GROUP	SIBLINGS OF INDEX CASES						PARENTS OF INDEX CASES					
	All siblings			All manifest tuberculosis cases		Ratio of tuberculosis deaths to all deaths	All parents			All manifest tuberculosis cases		Ratio of tuberculosis deaths to all deaths
	Total number	Living	Dead	Living	Dead		Total number	Living	Dead	Living	Dead	
						per cent						per cent
0- 4	147	—	147	—	—	—	—	—	—	—	—	—
5- 9	21	6	15	—	2	13.3	—	—	—	—	—	—
10-14	42	32	10	2	4	40.0	1	—	1	—	—	—
15-19	104	74	30	6	25	83.3	1	—	1	—	—	—
20-24	130	112	18	16	17	94.4	2	—	2	—	—	—
25-29	138	122	16	13	10	62.5	19	—	19	—	11	57.9
30-34	131	119	12	15	10	83.3	15	—	15	1	4	26.7
35-39	85	79	6	6	2	33.3	34	10	24	6	12	50.0
40-44	44	38	6	6	4	66.7	58	48	50	10	18	36.0
45-54	56	50	6	2	4	60.7	165	88	77	6	21	27.3
55-64	22	16	6	—	—	—	197	139	58	15	4	6.9
Over 64	10	10	—	—	—	—	156	94	62	2	4	6.5
	930	658	272	66	78	28.7	688	379	309	40	74	23.9

more essential than any technicality of its application.

Consistency in the correction of all group differences in age distribution has been obtained in this survey by the use of the Abridged Weinberg Method.[30] This method relates the absolute morbidity figures to the total number of persons who have survived a given period of manifestation, increased by one-half of those persons who at the time of the study were still within the age limits of this period. The resulting morbidity rates are average expectation figures valid for persons above the chief manifestation period, which in reinfection tuberculosis was conservatively assumed throughout this study to extend from the age of fifteen to twenty-nine.

An extension of the manifestation period in reinfection tuberculosis, which occurs mainly before the age of thirty, would naturally enlarge the difference between absolute and corrected morbidity rates, since the given statistical rate of reference includes only one-half of the persons observed in this period and omits entirely the persons who are below the age of fifteen, regardless of whether they are deceased or living. An over-extension might even be expected to affect the statistical accuracy, since the usefulness of the Abridged Weinberg Method depends no less on a relatively uniform distribution of the persons observed within the accepted period of manifestation than it depends on a representative size of the total material studied.

The use of this method in determining an estimated tuberculosis morbidity rate for a general population, which in age and racial distributions is comparable to that of the State of New York, yields a corrected rate of 1.37 per cent, while the absolute figure is 1.08 per cent (Table III). Both figures have been obtained by multiplying by ten for the white and by five for the non-white the present age-specific annual death rates as compiled by the U. S. Bureau of the Census. The use of a higher multiplier for whites than for non-whites has been considered necessary because the generally more rapid and destructive course of the disease in the negro may be expected to result in a lower ratio between annual morbidity and mortality figures in the non-white than in the white population.*

Of course, this estimated general morbidity rate is only a crude maximum figure for a population consisting of five-sixths whites and one-sixth non-whites. It does not compare with general morbidity rates obtained by direct sampling methods, although it may be used in the absence of more accurate morbidity figures for unselected population groups. It may be possible later on to secure a more exact general population rate for this survey by the subsequent inclusion of both the siblings of the marriage partners and the step-siblings of the index cases.

Meanwhile, however, there seem to be no valid reasons against using a general morbidity figure of about one per cent for the particular comparative purposes of the present study. The differences in the collective morbidity rates for the various groups of the given material are so pronounced that their statistical significance appears unquestionable and certainly independent of any possible variations in the general morbidity figure used for comparison.

(To be concluded)

*To explain this statistical procedure in more detail, one may say that it attempts to estimate the incidence of morbidity from the incidence of mortality, since direct data are available only for mortality at the present time. It is known that of two groups of whites and negroes developing tuberculosis in a given year, the negroes will show a higher proportion of deaths. Consequently, in estimating the incidence of morbidity one has to multiply the negro death rate by a figure which is lower than the one used in the white rate. The multiplier accepted in the study is one-half of the figure used in the white rate.

TRIPLOID VARIETIES OF CITRUS*

C. A. KRUG† AND OSWALDO BACCHI
Instituto Agronômico, Campinas, São Paulo, Brazil

THE citrus industry is of growing economic importance in the State of São Paulo, Brazil, as it plays an important role in the diversification of agriculture. Among the citrus varieties under cultivation, the "Baía" and "Baianinha," both similar to the "Washington Navel" orange, and several varieties of grapefruit are the most important. The fruit was shipped in pre-war times to Europe, mainly to England; and there are still considerable exports to the Argentine Republic. Home consumption of citrus fruits is increasing and growers hope to overcome the present difficulties incident to the discrepancies of the export market by producing citrus oil in large quantities.

Since 1937 various government agencies in the State of São Paulo have been engaged in a cooperative project of citrus investigations. These are conducted in the Division of Citrus Plants, Genetics and Cytology of the Instituto Agronômico at Campinas, in the Citrus Experiment Station of the same Institute at Limeira, and in the Genetics Division of the Agricultural College at Piracicaba. Those projects comprise numerous investigations on taxonomy, physiology, embryology, cytology, genetics and breeding of citrus fruits.

As part of the program of the Cytology Division it was decided to make a detailed study of the chromosomal constitution of all *Citrus* species and varieties grown at the Citrus Experiment Station at Limeira. In a preliminary article,[1] the junior author has already given an account of his determinations. As expected, it was found that most types had a chromosome constitution of $2n = 18$, the basic chromosome number of *Citrus* being $n = 9$, as had been determined previously by Frost,[2] Longley,[7] Kandelaki,[5] Lapin[6] and Nakamura.[11] One variety, however, the "Tahiti lime" was found to be triploid ($2n = 27$) and another, yet unidentified, to be a hypertriploid ($2n = 28$). In the progeny of the latter, two plants were found with 33 and 21 somatic chromosomes respectively.

In the spring of 1941 the senior author made further cytological investigations at the Citrus Experiment Station, Riverside, California, where he found one more triploid variety. New determinations of chromosome number in progenies of triploids and investigations on the meiosis of the same triploids were made at the Instituto Agronômico at Campinas after the publication of the above mentioned paper by Bacchi. In the present article all information so far available regarding triploid *Citrus* types will be presented.

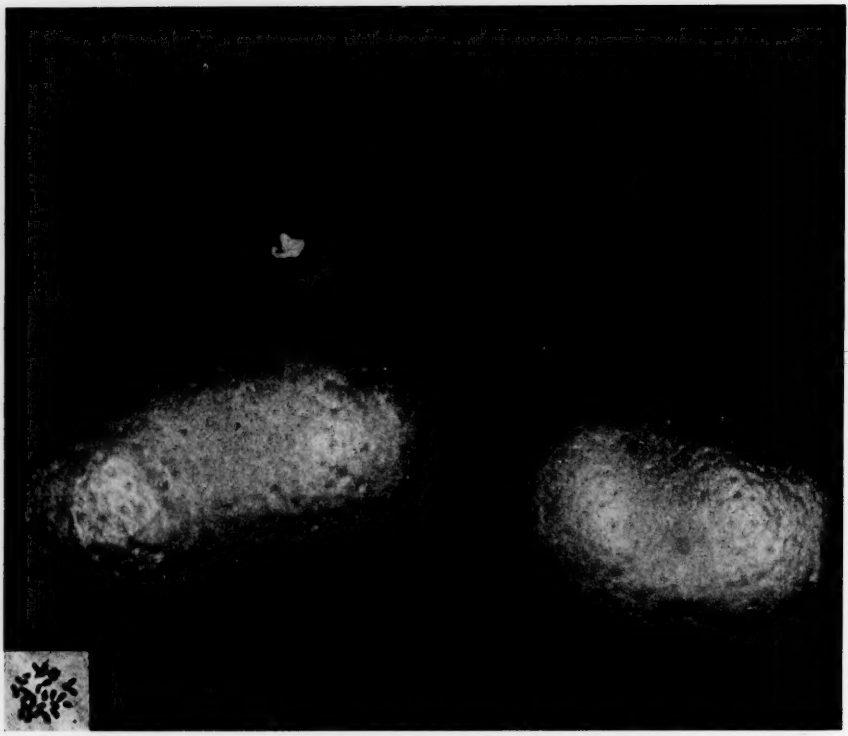
Triploidy and Hypertriploidy in *Citrus*

Review of Literature

With the exception of the findings by Bacchi,[1] triploidy in *Citrus* has only been found in artificial hybrids. The first artificial triploid citrus fruit was made deliberately by Swingle by pollinating *Fortunella hindsii* an extremely fruitful kumquat growing wild in the mountains of southeastern China (shown by Longley[7] to be a tetraploid) with a diploid limequat (*Citrus aurantifolia* × *Fortunella*

*Cytological observations of Citrus II, Instituto Agronômico series.

†The senior author wishes to express his appreciation to the John Simon Guggenheim Memorial Foundation, which enabled him to spend several months at the Citrus Experiment Station, Riverside, California, where part of the investigations were carried out; to the director and staff members of the Plant Breeding Division of that Station for all facilities offered. The authors also wish to express their thanks to A. J. T. Mendes and F. Juliano Filho of the Cytology Division of the Instituto Agronômico for general assistance during the work.



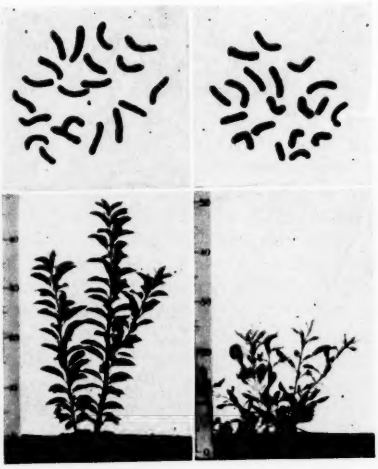
A TRIPLOID LIME

Figure 2

Leaves and fruit of the Tahiti lime, the only triploid citrus variety (with the exception of the derivative Bearss seedless lime) thus far found in cultivation. Like the Bearss, the Tahiti lime is also virtually seedless, though this appears not to be due primarily to its triploid nature. The insert (lower left) shows a microphotograph of the metaphase chromosomes. Twenty-seven chromosomes can be counted in contrast to the 18-20 chromosomes found in diploid forms.

margarita). Three hybrids resulting from this cross were found by Longley[8] to be triploids as expected; they are vigorous plants called procimequats which flower and fruit freely when only a few years old (Swingle[17]). Longley was especially interested in the cytological constitution of the hybrid and in the behaviour of the chromosomes at meiosis. At first metaphase 13 bivalents and a univalent were found. Sometimes the chromosomes were clumped together or more univalents were present at diakinensis; it was often not possible to distinguish between bivalent and trivalent chromosomes. The tetrads were usually abnormal, often with five and six spores, the mature pollen being abnormal. Frost[3] mentions the occasional production of triploid hybrids by crossing diploid citrus varieties, due to doubling of chromosome number probably during the formation of the nucleus. Trivalents are usually formed at the first meiotic division, together with bivalents and univalents; due to these irregularities, the microspores from each microsporocyte are variable in size and number.

Lapin[6] reports regarding the cytological constitution of 6524 citrus seedlings from his breeding projects, among which a total of 118 polyploids was



TAHITI SEEDLINGS

Figure 3

Seeds of the Tahiti lime average less than one-half per fruit and of these only a small fraction ever germinate. These two seedlings are diploids carrying extra chromosomes. That on the left has a diploid number of 20; the seedling on the right of 19 (the drawings are × 3000).

found, including triploids, tetraploids, one hypertriploid and one hexaploid. The appearance of the triploid seedlings is explained by the production of $2n$ pollen grains or of unreduced egg cells; in the cross *C. limon* × *C. paradisi* as many as 6.67 per cent triploids were found. The hypertriploid ($2n = 28$) was a hybrid between two varieties of lemon (*C. limon* × Meyer Chinese lemon). It is of interest to note that Luss[9] also reports the finding of a hypertriploid plant with *28* somatic chromosomes derived from a cross between the Novoafon lemon and the Meyer Chinese lemon, probably the same used by Lapin[7] in his hybridization work. Unfortunately nothing is known regarding the cytological constitution of this Chinese lemon, used as a male parent by both authors.

In the next section triploidy in *Citrus aurantifolia* Swingle will be discussed in detail.

Description of Citrus Triploids

Citrus aurantifolia Swingle

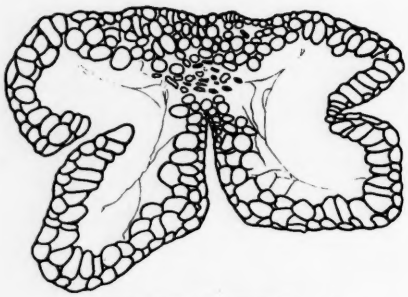
According to Hume[4] and Webber[23] this species of *Citrus* comprises a group of plants which is extremely variable, mainly with respect to fruit characters. It includes types with acid and sweet juice, the first group having either tight or loose rind. The "true limes" include only a few varieties of the so-called "Mexican" group, which produce small, yellow, seedy fruits with acid juice. Next in economic importance, comes the group of large fruited limes, in which the Tahiti and the Bearss varieties are best known for their seedlessness and otherwise favorable characters.

The Tahiti Lime

Origin and general morphological characters. This variety has been described in detail by Hume[4] and by Webber.[23,24,25] It is supposed to have originated from seed from ordinary limes imported into the United States from Tahiti.

At the Limeira Experiment Station the trees of this variety, derived from the plants imported from the U. S. A. tend to have a broad rather than erec habit of growth. They have abundan dark green leaves with very small wings on their petioles; the spines are small. Productivity is very satisfactory. The flowers are normal with the exception of the white or yellow anthers which do not dehisce, pollen being completely absent.[10,20] The fruits shown are typical of this variety (Figure 2), being broadly oval with an average length of 68 mm. They are very juicy, their rind color being lemon yellow; the style is persistent Most fruits are completely seedless; examining a total of 100 fruits at the Limeira Experiment Station, only 34 seeds were detected. A similar deficiency of seeds was found by Moreira and Gurgel,[10] i.e., between 0.2 and 0.4 seeds on an average per fruit.

Meiosis and cause of seedlessness. The seedlessness of this variety has been discussed by several investigators. Uphof[20] advanced the opinion that the lack of normal pollen grains and embryosacs is due to an early disintegration of the



EMPTY ANTHERS

Figure 4

Cross-section of mature anther of the Tahiti lime (× 100). A small amount of disorganized tissue is visible but no pollen grains.

microsporocytes and megasporocytes, which apparently do not divide; he compared this situation with the one found by Osawa[13] in the Washington Navel orange and the Satsuma orange (Unshiu mandarin). Torres[18] also noted the lack of pollen in this variety, and related this fact to the seedlessness of the variety; no cytological studies were made. This author[19] pollinated this variety with pollen of the Rough Lemon, but no seeds were obtained. Moreira and Gurgel[10] also did not succeed to obtain viable seeds by hybridizing the Tahiti lime and claim that seeds from uncontrolled pollinations did not germinate. Nakamura[11,12] who made extensive chromosome counts in the genus *Citrus*, classed the Tahiti lime as a normal diploid form, stating that the pollen-mother-cells degenerate at an early stage of development, as happens in the Washington Navel orange. Finally Bacchi[1] was the first to demonstrate that this variety of *Citrus aurantifolia* Swingle is a triploid ($2n = 27$; Figure 2). He advanced the suggestion that its cytological constitution might be responsible for its female and male sterility; no observations were made on meiosis.

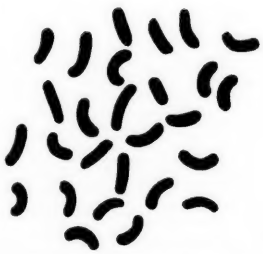
While determining the chromosome number of various species and varieties of *Citrus* at the Citrus Experiment Station, Riverside, California, the senior author confirmed Bacchi's results. A typical specimen of the Tahiti lime (C.E.S. 391; 3A-23-3) grown in the collection of that Experiment Station was determined by chromosome counts in root tips of cuttings to be triploid.

These results are not in accordance with the findings of Nakamura,[11] who classed this variety as a diploid ($2n = 18$). We might assume that the variety he examined was not a true Tahiti lime.

We believe that this variety probably originated as a seedling of one of the diploid limes. It is reasonable to assume that the triploid originated as a consequence of the fusion of one unreduced female or male gamete with a normal haploid gamete.

In order to determine whether the seedlessness of this triploid is due to its cytological constitution or to an early, probably genetically, controlled disintegration of microsporocytes and macrosporocytes, flower buds of various sizes were examined both in aceto-carmine smears and in microtome sections. This material was collected both at the Limeira Experiment Station (São Paulo, Brazil) and also at the Citrus Experiment Station, Riverside, California. Smears of anthers from very young buds revealed the presence of small microsporocytes, but not a single one was found to be dividing; older anthers were completely empty or contained a very limited number of mature pollen grains which were variable in size. Cross sections and longitudinal sections of mature anthers revealed that they are either completely empty or that they contain a small amount of usually disorganized tissue (Figure 4).

Based on these observations and on the results obtained by Uphof,[20] it is concluded that the seedlessness of this variety is not due to its triploid nature, since normally no meiotic divisions occur at all, the microsporcytes and also megasporocytes degenerating at an early stage of development. As a few pollen grains were found, and considering also that gametic seedlings (not nucellar) were obtained from this lime, as will be noted in the next section, it is evident that a limited number of microsporocytes and megasporocytes escape disintegration,



CHROMOSOMES OF BEARSS SEEDLESS LIME

Figure 5

Here the $2n$ chromosome number is twenty-seven. Magnification of drawing $\times 3900$.

their metiotic divisions probably occurring as in other triploids.

Cytological constitution of its progeny. As no seeds from controlled pollinations were available from this triploid variety, 54 fruits from open pollinated flowers were collected in January 1940. These furnished a total of eight seeds—not a single one of which germinated. In June of the same year 33 additional fruits were examined which had 11 seeds. Two of these were eliminated and of the remaining nine, only six germinated. Of these, two died at an early stage of development, and only three of the survivers furnished good material (root tips) for chromosome determination. One has 19, and the other two respectively 20 and 21 chromosomes. Considering the lack of pollen of this variety and assuming, therefore, that normal nine-chromosome pollen from surrounding trees of other diploid varieties was responsible for the formation of these seedlings, one concludes that only megaspores with a chromosome number close to nine give rise to viable embryosacs in this triploid variety.

As expected, the plants of this progeny are variable in size, growth habit and leaf characters (Figure 3). It is interesting to note that not one of the examined seedlings is a nucellar one, all being of gametic origin as indicated by their chromosome number.

A similar behavior has been noticed by Osawa[13] with the diploid Washington Navel orange, whose microsporocytes and megasporocytes usually degenerate in the tetrad stage producing no pollen. This author has also shown that occasionally normal embryosacs are formed, which might explain the occurrence of many seeds containing viable embryos, when its flowers are pollinated with pollen from other varieties, as recently found at the Limeira Experimental Station.[10] No information is so far available to indicate whether the seedlings derived from such seeds are of gametic origin or are of the nucellar type. It is possible that the development of the latter type is stimulated by cross pollination.

"Bearss Seedless" Lime

Origin and general morphological characters. According to Hume[4] this variety is "evidently a form of Tahiti," and Webber[23,24,25] states that it originated as a seedling of this variety, having similar characters, except that it has more upright growing branches and slightly smaller fruits. Detailed comparative studies were made by the senior author of both the Bearss and Tahiti varieties at the Citrus Experiment Station, Riverside, California, where they grow side by side. No appreciable differences in tree, leaf and fruit characters could be detected. It is very probable, therefore, that the Bearss is a nucellar seedling of the Tahiti lime. If so, these two varieties should be identical in cytological and genetical constitution.

Meiosis and cause of seedlessness. Apparently no cytological investigations have previously been made with this variety to determine the cause of its seedlessness. Leafy cuttings of one plant of this variety (CES-564; 3A-26-3) were therefore rooted in sand at the Citrus Experiment Station, Riverside, California, in the spring of 1941. Chromosome counts made in root tips revealed that this variety is also of triploid constitution ($2n = 27$; Figure 5). This finding constitutes a further indication that this variety originated as a nucellar seedling of the Tahiti.

Flower buds of several sizes collected

from the above mentioned tree were studied in aceto-carmine smear preparations and the same results were obtained as observed for the Tahiti Lime. In a few anthers groups of small, apparently normal, microsporocytes were detected. In other preparations abnormal and shrunken ones were found. No meiotic divisions could be seen, in spite of the fact that in mature anthers a few small degenerated pollen grains were present.

It is concluded that the cause of its seedlessness is therefore genetical rather than cytological, as observed for the Tahiti Lime, from which it is assumed to have originated.

Importance of Triploidy in Citrus Breeding

Up to the present date polyploidy has not been of particular importance in citrus breeding. New commercial varieties have arisen either by somatic variations, which are easily propagated through budding, or by hybridization. The latter process has not been very successful when closely related varieties within a species or between similar species have been crossed as pointed out by Frost.[3] Hybridizations between distantly related species and species belonging to distinct genera have shown in many cases great vigor and high fruitfulness as in tangelos, citranges, limequats, etc., produced by Webber and Swingle,[25] Webber,[22-24] Swingle, Robinson and Savage.[15] Some trigeneric hybrids are vigorous and fruitful, among them the citrangequats, described by Swingle and Robinson.[16] According to Frost[3] crosses within *Citrus nobilis* Lour. have been successful in producing new varieties of commercial importance.

With regard to polyploidy in citrus breeding, it might be stated that tetraploids are usually inferior to diploids. Their growth rate is reduced, the trees being smaller and less erect; they are usually less productive, with fruits of less commercial value due to thicker rind and more fibrous internal structure. Triploids originated by hybridization have been described to be similar in appearance with tetraploids; most seem to be rather unfruitful, their fruits being either seedless or having a smaller amount of seeds than most diploids.[3]

The Tahiti and the Bearss limes, described in the present paper, seem to be the first triploid citrus varieties of economic importance so far discovered. In spite of their cytological constitution, they are highly productive, their fruits being larger than ordinary limes from which they derived. They are very juicy and have a thin rind, besides being almost completely seedless. Considering their superior qualities and assuming that like other triploids these limes would be virtually seedless even though their microsporocytes and megasporocytes should not disintegrate, it is thought that their discovery is of special importance to citrus breeders. The synthesis of new triploids should therefore be encouraged, as it has now been demonstrated that these might be of great economic importance.

Summary

A review is presented of citrus cytology and account is given of the author's investigations of this subject. Two triploid citrus varieties are described which are of economic importance. It appears probable that one of these is a nucellar seedling derived from the other. The morphological and cytological characteristics of these triploids are given in detail.

Attention is called to the importance of triploidy in citrus breeding.

Literature Cited

1. Bacchi, O. *Jornal de Agronomia* 3(4): 249-258. 1940.
2. Frost, H. B. *Jour. Wash. Acad. Sc.* 15: 1-3. 1925.
3. ————. *Current Science,* Bungalore, India. Special Number on Genetics. pp. 24-27. 1938.
4. Hume, H. H. The cultivation of Citrus fruits. Macmillan Corp., New York. 1938.
5. Kandelaki, G. V. *Trav. Int. Bot. Tbilissi* 4:109-120. *Cit. Plt. Breed. Abstr.* 9:465. 1938.
6. Lapin, W. K. *USSR All Union Scient. Res. Inst. Humid Subtrop.* 1(4):1-68. 35 pp. 1937.
7. Longley, A. E. *Jour. Nat. Acad. Sc.* 15: 347-351. 1925.

8. ————————. *Jour. Wash. Acad. Sc.* 16: 543-545. 1926.
9. Luss, A. I. *Soviet Subtropica* No. 11 (15) :17-27. (*Cit. Plt. Breed. Abstr.* 6(4): 1372. 1935.)
10. Moreira, S., and J. T. A. Gurgel. *Bragantia* 1:669-711. 1942.
11. Nakamur, M. *Bull. Kagoshima Coll. Agric. and Forestry* 1:11-14. 1934.
12. ————————. *Studia Citrol.* 6:162-178. 1934.
13. Osawa, V. *Journ. Coll. Agric. Univ. of Tokio* 4:83-116. 1912.
14. Swingle, W. T., and H. J. Webber. *Yearbook of the U. S. Dept. Agric. for 1897.* 383-420. 1898.
15. Swingle, W. T., T. R. Robinson, and E. M. Savage. *U. S. Dept. Agric. Circular 181.* 1931.
16 Swingle, W. T., and T. R. Robinson. *Jour. Agric. Research* 23:229-238, 5 pls. 1923.
17. Swingle, W. T. The Citrus Industry, Vol. I, Chapt. IV. pp. 129-474. In Press. University of California Press. 1943.
18. Torres, J. P. *Philip. Jour. Agric.* 3 (3) :217-229. 1932.
19. ————————. *Philip. Jour. Agric.* 9 (2) :161-176. 1938.
20. Uphof, J. C. Th. *Gartenbauwiss.* 4 (6) :513-520. 1931.
21. Webber, H. I. *Yearbook U. S. Dept. Agric. for 1905.* 275-290, pls. 17-20. 1906.
22. ————————. *Yearbook U. S. Dept. Agric. for 1906.* 329-346, pls. 17-21. 1907.
23. ————————. Mimeogr. Paper of Citrus Exp. Sta., Riverside, Calif. 1940.
24. ————————. The Citrus Industry, Vol. I, Chapt. III. XX X pp. University of California Press. 1943.
25. Webber, H. G., and W. T. Swingle. *Yearbook of the U. S. Dept. Agric. for 1904.* 221-240. 1905.

◆

Citrus Chromosome Numbers

An exhaustive survey of the literature dealing with citrus chromosomes, and considerable new material of his own, is presented by Dr. C. A. Krug in the *Botanical Gazette* (Vol. 104, pp. 602-611, 1943). The orange subfamily, Aurantioideae, comprises a varied lot of botanical material, ranging in size from small shrubs to large trees. The smallest fruits are currant-like berries, which bulk very tiny compared with giant pommelos weighing several pounds. The ecological habits cover almost as wide a range, wet tropics, hot and cold deserts, and the temperate zone to the edge of the arctic. The basic chromosome number of this very diverse botanical material is with remarkable constancy nine. The chromosomes are similar in appearance and numerous hybrids between some extremely unlike forms are reported. This suggests that the chromosomal relationship between the citrus relatives is much closer than would be expected by comparing their taxonomic differences. There is a striking contrast between the uniformity of the chromosome number characterizing the citrus relatives and the polyploid series encountered in other plant genera such as the roses. Among the Aurantioideae remarkably few polyploid forms are encountered (only one up to now found growing wild)—a smattering of triploids in the genus *Citrus,* and tetraploids in cultivated species in four other genera.—R. C.

◆

A Quadruplet Set Consisting of Two Pairs of Identical Twins

PROFESSORS Gardner and Newman (Jour. Heredity, October, 1942) have commented on the rarity of the "theoretically possible type of two-egg quadruplets consisting of two pairs of one-egg twins." In view of the extreme rarity of quadruplet births, it appears that the single case of the Derner Quadruplets, which have been identified as "two pairs of one-egg" twins," should have been mentioned. The Derner quadruplets were born at Beuthen, Germany, on December 19, 1927. The diagnosis was made at the Kaiser Wilhelm Institute for Anthropology, Dahlem, Berlin, where the present writer had the opportunity of seeing the diagnostic data. I have unfortunately with me now no other data than the blood groups and types of the quads. They are as follows: I and II—*O, MN;* III and IV—*O, M.* The quads are females and a photograph has been published in Reinhold Lotze's *Zwillinge* (Oehringen, 1927), page 24.

S. S. Sarkar
Bose Institute, Calcutta

AN ISOLATED HUMAN CASE OF MALFORMED UPPER EXTREMITIES AND THORAX

EUGENE BROWN*
Department of Zoology, University of Georgia, Athens

THE literature covering congenital skeletal malformations and their possible heritability has already assumed formidable proportions. However, none of the general reviews or bibliographies with which the writer is familiar (Bagg,[1] Krogman,[3] Lewin,[4] Mall,[7] Milles,[8] Murphy,[9] Stocks and Barrington[12]) makes mention of the occurrence in one individual of such an assemblage of skeletal (and accompanying muscular) anomalies as is exemplified in the case here described. Rodenstein[10] and McConnell[6] have reported instances of phocomelus (of upper extremities only), or dolphin arms. But so far as the writer has been able to ascertain, the only published account of a case that is quite similar to the present one is that of Stiles and Dougan[11] (individual III-18 in their pedigree). Inasmuch as there had been a rather frequent occurrence of appendicular skeletal abnormalities in members of the family discussed by these two authors, the present report of an isolated case in at least five generations of a rather fertile family may not be without genetic interest.

Anatomical Description

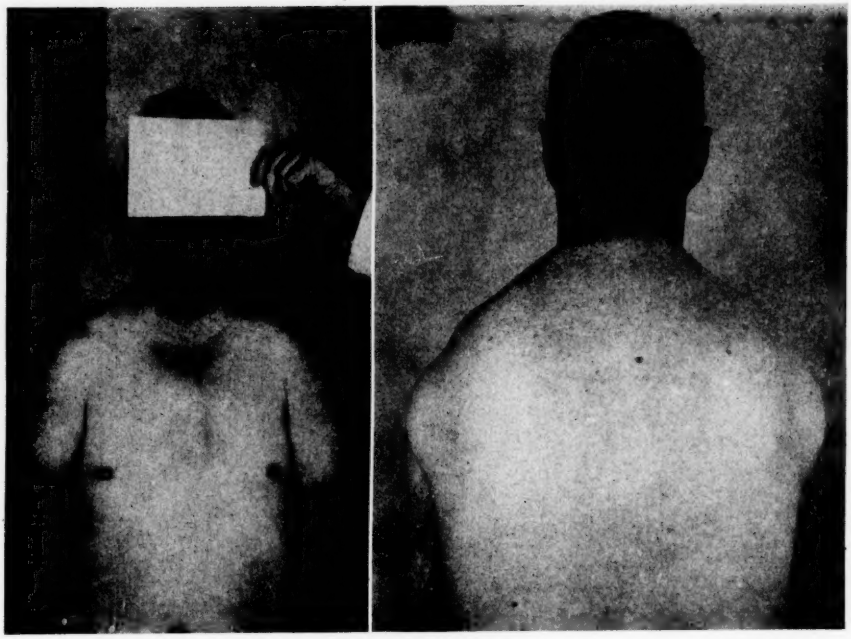
The present case is that of a 49 year old white man, six feet, 1½ inches tall, weighing 183 pounds. He is a resident of a small town in northeast Georgia, where he makes his living by serving, very capably, as county highway commissioner.

The most striking feature of the man's appearance is that, in the normal standing position, his fingers extend to a level about halfway between the nipples and the umbilicus, and the hands are permanently pronated and flexed (Figure 6). The arm and forearm are nine inches in length, the hand and fingers, seven. There are but three fingers on each hand. As is well seen from a dorsal view (Figure 6), the left arm is held more laterally than is the right. The downward slope from neck to tip of shoulder is markedly pronounced .

X-ray studies reveal anomalies in ribs, clavicles, scapulae, humeri, radii, ulnae, carpals, metacarpals and phalanges. The head of the first right rib articulates with the centra of what are apparently the first and second thoracic vertebrae, instead of with only the first vertebra (Figure 7*A*). The head of the second right rib articulates with only the centrum of the second thoracic vertebra. The clavicles, especially the left one, are markedly bowed upward, and their lateral ends extend over and beyond the acromion processes of the scapulae.

Only the head of the humerus appears to be present, and it is ankylosed to the glenoid region of the scapula (Figure 7*A*). The elbow joint, if such it may be called, is found just distal to the humeral head. As seen in Figure 7*B*, in the greater part of the "forearm" only one bone seems to be present. For this there are two possible interpretations. Either there has been an almost complete radio-ulnar synostosis, or else either the radius or ulna is, for the most part, absent. The latter view is probably the more tenable. At the proximal end (Figure 7*B*) it is seen that two bony elements are present.

*The writer gratefully acknowledges financial assistance received from the Department of Zoology of the University of Georgia, and favors from the staff of the X-ray laboratory of the Emory University Hospital.



SHORT ARMS

Figure 6

The extremely short arms of this individual and the three fingered hands are the most noticeable of a remarkable constellation of defects. The individual is of normal physical stature and of above average intelligence.

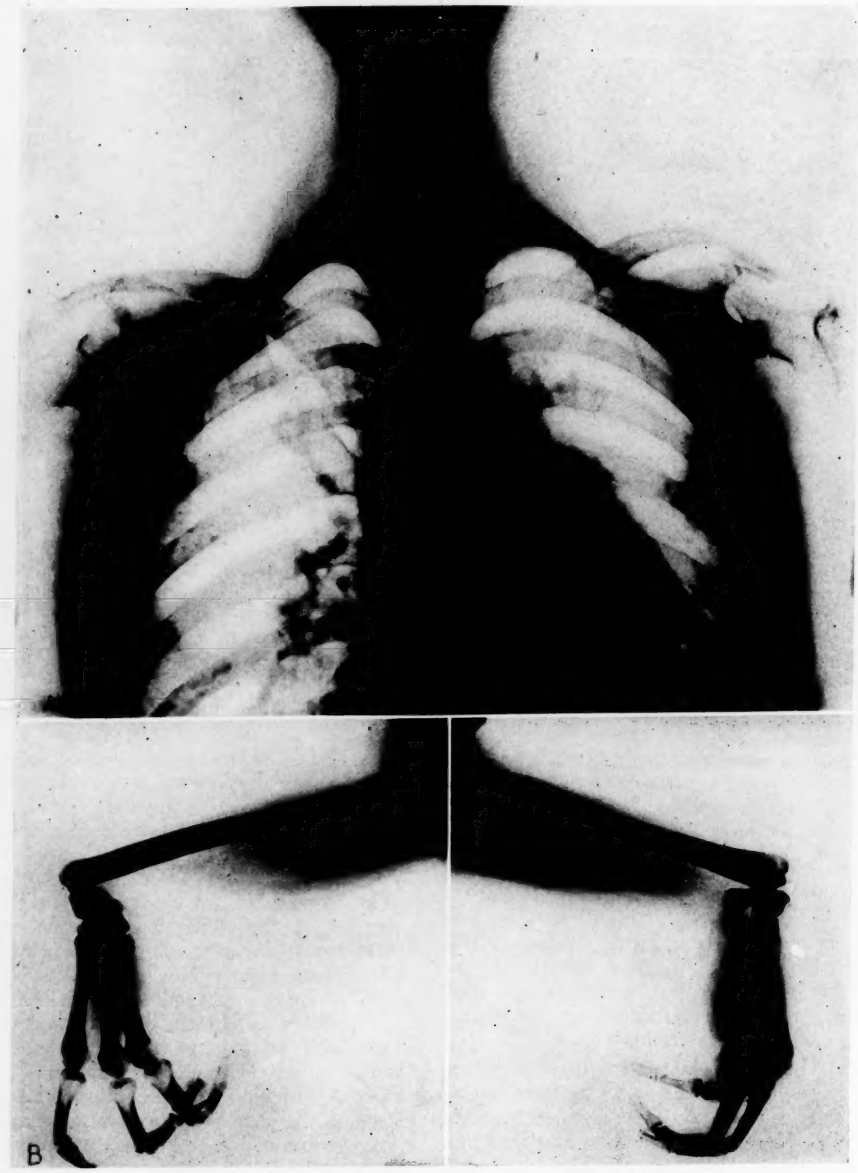
Neither resembles the proximal end of a normal radius or ulna. At the distal end (Figure 7*B*) a rudimentary bone is present, the well-formed bone clearly resembling a radius. Apparently the primary ossification center of the ulna was either never formed, or else it subsequently dedifferentiated.

The carpal bones are fused together to such an extent as to be unrecognizable (Figure 7*B*). The mass seems to be divisible into at least three portions but, as each of these probably represents parts of two or more elements of the normal carpus, no attempt has been made to assign names to them.

The metacarpals are three in number and normal in appearance. The phalanges of each of the three fingers are normal in number and appearance, except that the distal phalanx is hooked ventrally (Figure 7*B*).

Range of Movement

The "arms" are capable of being fully extended to the vertical position, the movement taking place (it is believed) entirely within the "elbow" joint. Vertical abduction of the "arm" is not possible nor, of course, is supination of the "forearm." Both flexion and extension of the "elbow" occur. The wrist joint, at rest, is flexed. Flexion can be continued until the palmar surface of the hand touches the anterior, or ventral, surface of the "forearm." Radial flexion is complete, but ulnar flexion is not allowed. Complete extension of the hand, either active or passive, is impossible. Both abduction and adduction of the fingers may occur. Neither of the lateral (anatomically speaking—always medial in this individual) fingers can be extended (probably due to abnormally short flexor tendons).



SKELETAL DEFECTS OF TRUNK AND APPENDAGES

Figure 7

This is a photograph of trunk (*A*) and arms (*B*) of individual showing remarkable syndrome of bilateral skeletal defects. Anomalies of the ribs, collar-bones, shoulder-blades, arm and hand bones, total up to a record number for one individual. That the defects are displayed on both sides and that they resemble cases of known hereditary defect are cogent reasons for considering that this isolated case may have a genetic basis, representing either a mutation or the segregation of a recessive gene.

Pedigree

The family history was obtained by the writer from the eldest sister of the propositus, she having some interest in genealogy. The propositus is sixth in a progeny of nine children, all the others normal. His father was one of six sibs, all normal. His mother was one of seven normal sibs. The pedigree consists of seventy-two individuals (including thirty nephews and nieces and sixteen grandnephews and grandnieces of the propositus spanning five generations). All save the propositus are normal with respect to appendicular skeleton. Since the sister made no attempt to conceal one case of mental deficiency and another of cleft palate, it is believed that her answers to questions were truthful.

Discussion

As to the possible inheritability of the condition, it seems likely that any such bilaterally symmetrical deformity would be the result of a widely generalized developmental mechanism such as might be expected to be present if the condition were hereditary. The pedigree reported by Stiles and Dougan[11] is a convincing argument in favor of the view that such malformations may be hereditary. The report of a first occurrence of this kind in a pedigree does not, of course, refute this possibility.

On the basis of their study Stiles and Dougan consider that the mode of inheritance of malformed upper extremities is one of "irregular dominance." They arrived at this conclusion because some individuals who were very probably heterozygous showed the trait (therefore it could not be due to a simple recessive factor), and because it appeared frequently in both sexes (therefore it probably was not due to a sex-linked factor*). Nor could it have been the result of a single dominant factor, because in one out of three cases the malformation skipped a generation.

In the writer's opinion nothing is gained by attempting to assign a name to a type of inheritance for which there are so little data. Moreover, since there was such a wide degree of variation in the extent to which the persons in the pedigree of Stiles and Dougan were deformed (some had only malformed fingers, some malformed wrists and fingers, and others malformed shoulders, arms, wrists and fingers), one is not justified in treating the data as if there were only two classes—normal and malformed. In all probability many different genetic factors are involved, perhaps some concerned with irregular ossification patterns in general and others bringing about a localization of the anomalous developmental behavior. Obviously if such be the case, one cannot predict the outcome of given matings with any reasonable degree of certainty.

Attention might be called to the absence in the present case of any "progressive hereditary" tendency such as has been discussed by Goldschmidt[2] and others. Such a tendency did seem to be present to a certain extent in the family described by Stiles and Dougan.

The propositus was a bachelor at the time of the examination, but he has since married. However, the advanced age of his wife renders it unlikely that they will furnish any additional evidence as to the inheritability of the condition. This is pleasing to the eugenicist but somewhat disappointing to the more callous geneticist.†

Of course, one's task is not over when he concludes that a certain pathological or abnormal condition is hereditary. It still remains for him to discover the *modus operandi* of the genetic factors. But any attempted discussion of the etiology of the present case would have to be too speculative to be justified. Mall[7] and Murphy[9] have discussed the numerous factors which might conceivably play roles in the production of mon-

*They might also have mentioned an equally cogent reason, *viz.*, that an affected woman had four normal sons.

†The writer has learned of four other similar cases which he hopes to study as soon as funds for travel and for X-ray pictures become available.

strosities. According to Lewis,[5] as early as the fifth week of normal ontogeny the mesenchymal and cartilaginous anlagen of the humerus, radius, ulna, individual carpal bones and elements of the five fingers are recognizable. Therefore it seems reasonable to suppose that, whatever the cause (hereditary or otherwise) of the anomalous development in the present case, it had begun to operate at least as early as the fifth week after conception. Until more is learned of the physiological genetics of skeletal tissues, it is perhaps best for one to refrain from speculating on the probable cause or causes of this condition.

For the benefit of any subsequent investigators who may wish to seek correlations between the incidence of congenital malformations and such factors as are given below, the following data are given. The propositus was the sixth in order of birth, being born at full term, in the month of January. His mother was 32 years old at the time of his birth and his father was 33. The reproductive rate of the mother was high, there having been six pregnancies during the 134 months which elapsed between the time of conception of her first child and the birth of her malformed one. Her third child was stillborn (but normally formed) at seven months, and three normal children followed the birth of the abnormal child. Members of the family were positive that there had been no consanguineous marriages as far back as the grandparents of the propositus, and they were of the opinion that none had occurred for at least two generations preceding this.

Summary

1. The occurrence of an isolated case of congenital, bilateral malformed upper extremities in a five-generation pedigree is reported.

2. In addition to minor anomalies of the ribs, the clavicles and scapulae are misshapen, the humeri rudimentary, the shoulder joint apparently immobile, the ulna almost entirely absent, carpals fused together, and two fingers absent on each hand.

3. The following movements are not allowed: vertical abduction of "arm," supination of "forearm," extension of wrist, ulnar flexion of wrist, extension of lateral fingers.

4. The pedigree of the individual shows no other case of outwardly apparent skeletal anomaly, but the bilateral nature of the condition and its close resemblance to deformities known to be inherited, suggests that it might be the result of a generalized inheritable developmental mechanism. If so, many genetic factors are probably involved.

Literature Cited

1. Bagg, H. J. *Amer. Jour. Anat.*, 43(2): 167-221. 1929.

2. Goldschmidt, Richard. *Jour. Heredity*, 29:140-142. 1938.

3. Krogman, Wilton M. A bibliography of human morphology. 1914-1939. University Press, Chicago. 1941.

4. Lewin, Philip. *Amer. Jour. Roentgenology*, 4:431-448. 1917.

5. Lewis, W. H. *Amer. Jour. Anat.*, 1: 145-184. 1902.

6. McConnell, Guthrie. *Int. Assoc. of Med. Mus. and Jour. of Tech. Meth. Bull.*, 8:156-159. 1922.

7. Mall, Franklin P. *Jour. Morph.*, 19:3-367. 1908.

8. Milles, Bess Lloyd. *Jour. Heredity*, 19:28-46. 1928.

9. Murphy, Douglas P. Congenital malformations. Univ. Penn. Press, Philadelphia. 1940.

10. Rodenstein, L. A. *Amer. Jour. Obstetrics*, 8:663-664. 1875-1876.

11. Stiles, Karl A., and Paul Dougan. *Jour. Heredity*, 31:65-72. 1940.

12. Stocks, Percy, and Amy Barrington. Treasury of Human Inheritance, 3, pt. 1. 182 pp.

SOME INSTITUTIONS
ORDERING
JOURNAL LANTERN SLIDES
Connecticut College
Elizabethtown College
Emory University
Evander Childs High School, N. Y. City
Franklin and Marshall College
Grover Cleveland High School, N. Y. City
Harvard University
Independent School District, Virginia, Minn.
Pennsylvania State College
Purdue University
Rhode Island State College
Rutgers University
San Diego State College
Swiss Federal Institute of Technology
University of Pittsburgh
University of Southern California
University of Wisconsin
Williams College
Wilson College
A list of over 200 slides which may be ordered by number will be sent on request; or order from the Journal, giving volume, page, and figure number. Inquiries solicited regarding slides available to illustrate specific subjects. Price 75c each, $8.00 per dozen.
AMERICAN GENETIC ASSOCIATION
Victor Building
Washington, D .C.

OUR MEN NEED

★ BOOKS ★

SEND

ALL YOU CAN SPARE

GIVE A BOOST WITH A BOOK

Good books, in good condition, are wanted by the

1943 VICTORY BOOK CAMPAIGN

for men in all branches of the service. Leave yours at the nearest collection center or public library.

DEPENDABLE

. . . *feeds for your "Lab" Animals*

Dickinson's Globe Feeds are carefully prepared by a firm with over 88 years of experience . . . a pioneer in the manufacture of mixed animal feeds.

Globe Feeds supply complete balanced rations. Because they are dry, storing is easy and feeding is simplified with a great reduction in waste. One can always expect the same high standard of quality in Globe Feeds from year to year.

FOR RABBITS

Globe Rabbit Glob-ets to be supplemented with hay.
Globe Rabbit with Alfalfa, meal form—complete.
Globe Rabbit with Alfalfa, pellet form—complete.

FOR RATS AND MICE

Dickinson's Dog Food — Nugget — Pellet or meal form. Dickinson's Kibbled Dog Food.

FOR GUINEA PIGS

Globe Rabbit Glob-ets to be supplemented with hay.
Globe Rabbit with Alfalfa, meal form—complete.
Globe Rabbit with Alfalfa, pellet form—complete.
Supplement with Vitamin C carrier three times a week.

GENEROUS SAMPLES FREE

To supervisors of experimental animals, we will send a free sample, large enough to make an adequate test. Just tell us the kind and quantity of food desired.

THE ALBERT DICKINSON COMPANY

2714 WEST 35TH STREET **CHICAGO, ILL.**